15

PATENT COOPERATION TREATY

PCT

REC'D 24 NOV 2000
WIPO PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
R-896-WO		
International application No. PCT/GB99/03011	International filing date (day/month/year) 09/09/1999	Priority date (day/month/year) 10/09/1998
International Patent Classification (IPC) or national classification and IPC C12N15/82		
Applicant MONSANTO PLC et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 8 sheets, including this cover sheet.

   ☐ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the report
   - II ☐ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☒ Lack of unity of invention
   - V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations suporting such statement
   - VI ☒ Certain documents cited
   - VII ☒ Certain defects in the international application
   - VIII ☒ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
04/04/2000	22.11.2000
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized officer Chavanne, F Telephone No. +49 89 2399 8399

Form PCT/IPEA/409 (cover sheet) (January 1994)

I. Basis of the report

1. This report has been drawn on the basis of *(substitute sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to the report since they do not contain amendments (Rules 70.16 and 70.17).)*:
   Description, pages:

   1-56 as originally filed

   Claims, No.:

   1-25 as originally filed

   Drawings, sheets:

   1/56-56/56 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
   ☐ the language of publication of the international application (under Rule 48.3(b)).
   ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   ☐ contained in the international application in written form.
   ☐ filed together with the international application in computer readable form.
   ☐ furnished subsequently to this Authority in written form.
   ☐ furnished subsequently to this Authority in computer readable form.
   ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
   ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   ☐ the description, pages:
   ☐ the claims, Nos.:

☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

IV. Lack of unity of invention

1. In response to the invitation to restrict or pay additional fees the applicant has:

   ☐ restricted the claims.

   ☐ paid additional fees.

   ☐ paid additional fees under protest.

   ☐ neither restricted nor paid additional fees.

2. ☒ This Authority found that the requirement of unity of invention is not complied and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is

   ☐ complied with.

   ☒ not complied with for the following reasons:
   see separate sheet

4. Consequently, the following parts of the international application were the subject of international preliminary examination in establishing this report:

   ☒ all parts.

   ☐ the parts relating to claims Nos. .

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	14-17, 23-25
	No:	Claims	1-13, 18-22
Inventive step (IS)	Yes:	Claims	
	No:	Claims	1-25

Industrial applicability (IA)	Yes:	Claims	1-25
	No:	Claims	

2. Citations and explanations
   see separate sheet

VI. Certain documents cited

1. Certain published documents (Rule 70.10)

and / or

2. Non-written disclosures (Rule 70.9)

   see separate sheet

VII. Certain defects in the international application

The following defects in the form or contents of the international application have been noted:
see separate sheet

VIII. Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:
see separate sheet

IV. Lack of unity of invention

1. The present application lacks unity *a posteriori* (Rule 13.1 and 13.3 PCT) for the following reasons:

   The common inventive concept underlying the present application can be seen in the provision of nucleotide sequences encoding starch branching enzymes (SBE) from wheat. Two nucleotide sequences encoding starch branching enzymes from wheat have already been described in the prior art (see e.g. D1). As a consequence, this common concept no longer exists. Correspondingly, claims 1-25 are not linked by a single inventive concept. Therefore, claims 1-25 lack unity *a posteriori* (Rule 13.1 and 13.3 PCT). They relate to two different inventions, namely:

   Invention 1 (claims 1-4, 6 and 8, completely; claims 7 and 9-25, partially):
   Nucleotide sequence of wheat SBEII-1 genes, and functional equivalents thereof, the corresponding amino acid sequence, methods of altering the characteristics of a plant by introducing said nucleotide sequence in a plant, and plants having said altered characteristics.

   Invention 2 (claim 5 completely; claims 7 and 9-25, partially):
   Nucleotide sequence of a sub-class of wheat SBEII-2 genes, and functional equivalents thereof, the corresponding amino acid sequence, methods of altering the characteristics of a plant by introducing said nucleotide sequence in a plant, and plants having said altered characteristics.

V. Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Reference is made to the following documents:

   D1: Plant Physiol.
   Vol. 118, pp. 37-49, 1998

2. D1 describes the wheat SBEIIa-1 and SBEIIa-2 sequences, which correspond to two sub-classes of the wheat SBEII-2 genes. D1 describes the isolation and characterisation of the barley SBEIIa and SBEIIb genes (page 41). D1 also mentions the nucleotide sequence and the corresponding amino acid sequence of the maize SBEIIa and SBEIIb genes. Phylogenetic analysis shows that said sub-classes of the wheat SBEII-2 gene cluster with the barley and maize SBEIIa genes on a phylogenetic tree, and that the barley and maize SBEIIb genes cluster together on the same tree (figures 2 and 3).

3. The subject-matter of claim 1 refers to a nucleotide sequence encoding substantially the amino acid sequence of the wheat SBEII-1. Due to the term substantially, said claimed subject-matter encompasses nucleotide sequences encoding homologues of the wheat SBEII-1. Claim 1 also refers to functional equivalents of the claimed nucleotide sequence. Homologues of a gene can be seen as functional equivalents of said gene. Homologues of the wheat SBEII-1 genes, notably SBEIIb from wheat and barley, are known in the art (see e.g. D1). Thus, the subject-matter of claim 1 is not novel.
The subject-matter of claims 2-6 refers to a nucleotide sequence comprising the sequence of a wheat SBEII-1 or SBEII-2 sub-class, or a functional equivalent thereof. The subject-matter of claim 8 refers to a nucleotide sequence comprising the 3' untranslated region of subclasses of the wheat SBEII genes, or functional equivalent thereof. Homologues of a gene can be considered as functional equivalents of said gene. The SBEIIa and SBEIIb genes of barley and maize are homologues of the wheat SBEII-2 and SBEII-1 genes, respectively. Thus, the subject-matter of claims 2-6 and 8 is not novel.
Thus, in view of e.g. D1, due to the too broad formulation of claims 1-6 and 8, claims 1-13 are not novel.
Therefore, claims 1-13 do not meet the requirements of Article 33(2) PCT.

4. Due to the segregation during meiosis the progeny of transformed plants encompasses also non-transformed plants. Therefore, claims 18-22 do not meet the requirements of Article 33(2) PCT.

5. Should the applicant overcome the novelty objections on the above mentioned claims, claims 1-25 of the present application would still not be recognised as

inventive for the following reasons:

The closest prior art to evaluate the inventiveness of the present application is D1. The problem to be solved by the present application was to provide alternative wheat starch branching enzyme (SBE) genes.
The present application solves this problem by providing sequences of genes encoding sub-classes of the wheat SBEII-1 and SBEII-2.

The subject-matter of claims 5, 7 and 9-13 differs from the teaching of D1 in that the claimed SBEII-2 sequence correspond to a third sub-class of the SBEII-2 genes.
However, it is known in the art that the wheat genome has been triplicated throughout evolution. Thus, it is expected to find three copies of each gene, i.e. three sub-classes. Thus, the man skilled in the art, aware of the evolution of the wheat genome, in view of D1, which describes two sequences of the wheat SBEII-2 gene, would only need to apply common knowledge and commonly used standard methods (i.e. PCR amplification, hybridisation, etc...) to isolate and characterise the third sequence of said gene, and come to the subject-matter of claims 5, 7 and 9-13. Thus, these claims are not inventive.

The subject-matter of claims 1-4, 6 and 8 differ from D1 in that the claimed sequences of the wheat SBEII correspond to a different class (SBEII-1). However, in view of the phylogenetic analysis disclosed in D1 (figure 3), it appears that the wheat SBEII-2 genes are the wheat homologues of the barley and maize SBEIIa genes. It is known in the art that the barley and maize SBEII genes are related to each other (see D1) and form two different classes, SBEIIa and SBEIIb. Thus, in view of the close relationship between wheat, barley and maize shown in D1, it is expected that the wheat SBEII genes would also have a second class corresponding to the barley and maize SBEIIb. As mentioned above, the wheat comprises three homologous genomes. Thus, each class from wheat should be composed of three sub-classes. Isolation and characterisation methods of closely related genes are well-known in the art and commonly applied. Thus, the man skilled in the art, aware of D1, would not require any inventive skill to come to the subject-matter of claims 1-4, 6 and 8. Therefore, said claims are not inventive.

Known methods based on a non inventive product and the use of a non inventive product according to known methods are also not inventive. Thus, claims 14-25 are not inventive.

Therefore, claims 1-25 do not meet the requirements of Article 33(3) PCT.

VI. Certain documents cited

Certain published documents (Rule 70.10)

1. WO 99/14314

VII. Certain defects in the international application

1. The subject-matter of claims 18, 19 and 21 is not limited to one single product as it should be, but relates to four different ones: a plant, a plant cell, the progeny of a plant or part of a plant.

VIII. Certain observations on the international application

1. Claims 1-6 and 8 lack clarity due to the term "substantially". This term is not suitable to clearly define the scope of the claim, because it is without technical significance and its vagueness makes it entirely opened to individual interpretation. Claims 1-6 and 8 further lack clarity due to the expression "functional equivalent". In fact, said expression does not seem to be adapted in connexion with a nucleotide sequence, but would be adapted in relation with an amino acid sequence. Thus, claims 1-6 and 8 do not meet the requirements of Article 6 PCT.

2. The formulation "...comprising..." in claims 2-5 and 8 does not clearly define the scope of these claims. Thus, this expression should be replaced with "consisting of" (Article 6 PCT).

‥\TENT COOPERATION TRE‾‾Y

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF ELECTION

(PCT Rule 61.2)

To:

Assistant Commissioner for Patents
United States Patent and Trademark Office
Box PCT
Washington, D.C.20231
ETATS-UNIS D'AMERIQUE

in its capacity as elected Office

Date of mailing (day/month/year) 28 April 2000 (28.04.00)	
International application No. PCT/GB99/03011	**Applicant's or agent's file reference** HCM/C397.02/U
International filing date (day/month/year) 09 September 1999 (09.09.99)	**Priority date** (day/month/year) 10 September 1998 (10.09.98)
Applicant GOLDSBROUGH, Andrew et al	

1. The designated Office is hereby notified of its election made:

[X] in the demand filed with the International Preliminary Examining Authority on:

04 April 2000 (04.04.00)

[ ] in a notice effecting later election filed with the International Bureau on:

2. The election [X] was

[ ] was not

made before the expiration of 19 months from the priority date or, where Rule 32 applies, within the time limit under Rule 32.2(b).

The International Bureau of WIPO **34, chemin des Colombettes** **1211 Geneva 20, Switzerland** Facsimile No.: (41-22) 740.14.35	Authorized officer Olivia RANAIVOJAONA Telephone No.: (41-22) 338.83.38

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

To:

BOSCH, Henry
Monsanto Services International
Avenue de Tervuren 270-272
1150 Brussels
BELGIQUE

PCT

NOTIFICATION OF THE RECORDING OF A CHANGE

(PCT Rule 92bis.1 and Administrative Instructions, Section 422)

Date of mailing (day/month/year) 06 April 2000 (06.04.00)	
Applicant's or agent's file reference HCM/C397.02/U	**IMPORTANT NOTIFICATION**
International application No. PCT/GB99/03011	**International filing date** (day/month/year) 09 September 1999 (09.09.99)

1. The following indications appeared on record concerning:
[X] the applicant [ ] the inventor [ ] the agent [ ] the common representative

Name and Address	State of Nationality	State of Residence
PLANT BREEDING INTERNATIONAL CAMBRIDGE LIMITED Maris Lane Trumpington Cambridge CB2 2LQ United Kingdom	GB	GB
	Telephone No.	
	Facsimile No.	
	Teleprinter No.	

2. The International Bureau hereby notifies the applicant that the following change has been recorded concerning:
[X] the person [X] the name [X] the address [ ] the nationality [ ] the residence

Name and Address	State of Nationality	State of Residence
MONSANTO PLC P.O.Box 53 Lane End Road High Wycombe Buckinghamshire HP12 4HL United Kingdom	GB	GB
	Telephone No.	
	Facsimile No.	
	Teleprinter No.	

3. Further observations, if necessary:

4. A copy of this notification has been sent to:
[X] the receiving Office
[ ] the International Searching Authority
[ ] the International Preliminary Examining Authority
[X] the designated Offices concerned
[ ] the elected Offices concerned
[ ] other:

The International Bureau of WIPO **34, chemin des Colombettes** **1211 Geneva 20, Switzerland** Facsimile No.: (41-22) 740.14.35	Authorized officer Maria Victoria CORTIELLO Telephone No.: (41-22) 338.83.38

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF THE RECORDING OF A CHANGE

(PCT Rule 92bis.1 and Administrative Instructions, Section 422)

To:

BOSCH, Henry
Monsanto Services International
Avenue de Tervuren 270-272
1150 Brussels
BELGIQUE

Date of mailing (day/month/year) 06 April 2000 (06.04.00)	
Applicant's or agent's file reference HCM/C397.02/U	**IMPORTANT NOTIFICATION**
International application No. PCT/GB99/03011	**International filing date** (day/month/year) 09 September 1999 (09.09.99)

1. The following indications appeared on record concerning:

☐ the applicant ☐ the inventor ☒ the agent ☐ the common representative

Name and Address	State of Nationality	State of Residence
KEITH W NASH & CO 90-92 Regent Street Cambridge CB2 1DP United Kingdom	Telephone No. 01223 355477 Facsimile No. 01223 324353 Teleprinter No.	

2. The International Bureau hereby notifies the applicant that the following change has been recorded concerning:

☒ the person ☒ the name ☒ the address ☒ the nationality ☒ the residence

Name and Address	State of Nationality	State of Residence
BOSCH, Henry Monsanto Services International Avenue de Tervuren 270-272 1150 Brussels Belgium	Telephone No. 02 776 4133 Facsimile No. 02 776 4556 Teleprinter No.	

3. Further observations, if necessary:

4. A copy of this notification has been sent to:

☒ the receiving Office
☐ the International Searching Authority
☐ the International Preliminary Examining Authority
☒ the designated Offices concerned
☐ the elected Offices concerned
☐ other:

The International Bureau of WIPO **34, chemin des Colombettes** **1211 Geneva 20, Switzerland** Facsimile No.: (41-22) 740.14.35	Authorized officer Maria Victoria CORTIELLO Telephone No.: (41-22) 338.83.38

PCT

WORLD INTELLECTUAL PROPERTY ORGANIZ
International Bureau

INTERNATIONAL APPLI... ...ION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 7 :		(11) International Publication Number: WO 00/15810
C12N 15/82, 9/10, A23L 1/0522, A01H 5/00	A1	(43) International Publication Date: 23 March 2000 (23.03.00)

(21) International Application Number: PCT/GB99/03011

(22) International Filing Date: 9 September 1999 (09.09.99)

(30) Priority Data:
98307337.0 10 September 1998 (10.09.98) EP

(71) Applicant (*for all designated States except US*): PLANT BREEDING INTERNATIONAL CAMBRIDGE LIMITED [GB/GB]; Maris Lane, Trumpington, Cambridge CB2 2LQ (GB).

(72) Inventors; and
(75) Inventors/Applicants (*for US only*): GOLDSBROUGH, Andrew [GB/GB]; 50 Melvin Way, Histon, Cambridge CB4 9HY (GB). COLLIVER, Steve [GB/GB]; 1 Astwood Road, Bourne End, Cranfield, Bedfordshire MK43 0AU (GB).

(74) Agent: KEITH W NASH & CO; 90-92 Regent Street, Cambridge CB2 1DP (GB).

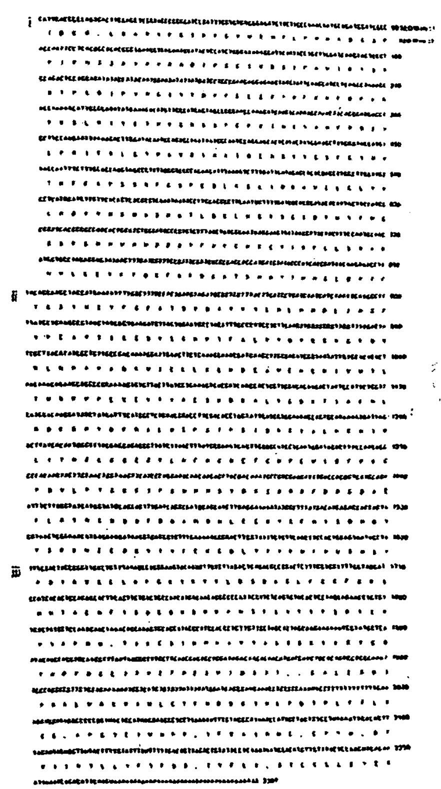
(81) Designated States: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) Title: ISOFORMS OF STARCH BRANCHING ENZYME II (SBE-IIA AND SBE-IIB) FROM WHEAT

(57) Abstract

A class of wheat SBEII genes, called SBEII-1, can be used to influence properties of starch produced by a plant, including the gelatinisation temperature of the starch. The starch is useful, eg. in bakery products.



FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece			TR	Turkey
BG	Bulgaria	HU	Hungary	ML	Mali	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MN	Mongolia	UA	Ukraine
BR	Brazil	IL	Israel	MR	Mauritania	UG	Uganda
BY	Belarus	IS	Iceland	MW	Malawi	US	United States of America
CA	Canada	IT	Italy	MX	Mexico	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NE	Niger	VN	Viet Nam
CG	Congo	KE	Kenya	NL	Netherlands	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NO	Norway	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	NZ	New Zealand		
CM	Cameroon			PL	Poland		
CN	China	KR	Republic of Korea	PT	Portugal		
CU	Cuba	KZ	Kazakstan	RO	Romania		
CZ	Czech Republic	LC	Saint Lucia	RU	Russian Federation		
DE	Germany	LI	Liechtenstein	SD	Sudan		
DK	Denmark	LK	Sri Lanka	SE	Sweden		
EE	Estonia	LR	Liberia	SG	Singapore		

ISOFORMS OF STRACH BRANCHING ENZYME II (SBE-IIA AND SBE-IIB) FROM WHEAT

Field of the Invention

This invention relates generally to plant starch compositions, and concerns novel nucleotide sequences; polypeptides encoded thereby; vectors and host cells and host organisms comprising one or more of the novel sequences; a method of altering one or more characteristics of a plant; a plant having altered characteristics; starch obtained from such plants; and uses of the starch.

Background to the Invention

The majority of developments in cereal science in the recent past have concentrated primarily on the functionality of the gluten protein sub-units and their role in bakery systems. This has been greatly facilitated by the abundance of natural variation between cultivators for the gluten protein sub-unit components.

In contrast, although flour from commercially grown wheat varieties contains approximately 75-85% starch, the role of starch from a breeding perspective has been overlooked; this is largely due to the difficulty of measuring differences in starch structure. Of the limited amount of work that has been carried out however, there appears to be a lack of natural variation between different wheat cultivars. With the advent of recombinant DNA and gene transfer technologies it is now possible to create new variation *in planta*, therefore directly modifying starch composition in wheat becomes a realistic target.

Starch is the major form of carbon reserve in plants, constituting 50% or more of the dry weight of many storage organs, e.g. tubers, seeds of cereals. Starch is used in numerous food and industrial applications. In many cases, however, it is necessary to modify the native starches, via chemical or physical means, in order to produce distinct properties to

suit particular applications. It would be highly desirable to be able to produce starches with the required properties directly in the plant, thereby removing the need for additional modification. To achieve this via genetic engineering requires knowledge of the metabolic pathway of starch biosynthesis. This includes characterisation of genes and encoded gene products which catalyse the synthesis of starch. Knowledge about the regulation of starch biosynthesis raises the possibility of "re-programming" biosynthetic pathways to create starches with novel properties that could have new commercial applications.

The most significant property of starch derives from the ability of the native granular form to lose its order and to swell and absorb water upon suitable treatment, thereby conferring viscosity and texture, in a process known as gelatinisation. Gelatinisation has been defined (W A Atwell *et al*, 1988) as "... the collapse (disruption) of molecular orders within the starch granule manifested in irreversible changes in properties such as granular swelling, native crystallite melting, loss of birefringence, and starch solubilisation. The point of initial gelatinisation and the range over which it occurs is governed by starch concentration, method of observation, granule type, and heterogeneities within the granule population under observation".

14 molecules of water per molecule of anhydrous glucose, i.e. a minimum of 75% water, are necessary for full starch gelatinisation (Donovan, 1979). Starch gelatinisation is usually caused by heat, but can be caused by physical damage and some chaotropic agents, mainly dimethylsulphoxide (DMSO), urea, calcium chloride, strong base and acid.

The various events taking place during gelatinisation can be followed by various methods, including birefringence, X-ray diffraction, differential scanning calorimetry (DSC), ^{13}C NMR. Swelling can be monitored by various methods, particularly rheology.

Differential scanning calorimetry (DSC) is a destructive method which records an endothermic event on heating of granules, generally thought to measure the temperature and the endothermic energy (delta H) required for the melting of the native crystallites. Starch gelatinisation temperature is independent of water content above 75% water (described as excess water), but increases when water is limited (Donovan, 1979).

The rate and extent of starch granule swelling upon heating dictate the type of viscosity development of aqueous starch suspensions on heating. Swelling behaviour is therefore of utmost technological importance. Viscosity increase on heating can be conveniently measured by a Brabender amylograph (Brabender is a Trade Mark) (Kennedy and Cabalda, 1991) or using a Rapid Visco analyser (Rapid Visco is a Trade-Mark from Newport Scientific, Australia). Figure 1 is a typical viscoamylgraph profile for wheat starch, produced in this way, showing changes in starch during and after cooking. As starch granules swell on uptake of water, in a process known as pasting, their phase volume increases, causing an increase in viscosity. The onset of pasting is indicated at A in Figure 1. Peak viscosity, indicated at B in Figure 1, is achieved when maximum phase volume is reached. Shear will then disrupt/cause fragmentation of the swollen granules, causing the viscosity to decrease. Complete dispersion is indicated at C in Figure 1. This has been confirmed by an oscillatory rheology study of starch pastes at various stages of the viscosity profile (Svegmark and Hermansson, 1990). The viscosity onset temperature and peak viscosity are indicative of the initiation and extent of swelling, respectively. On cooling, leached amylose forms a network in a process involving reassociation of molecules, or retrogradation, causing an increase in viscosity as indicated at D in Figure 1. Retrogradation (or set-back) viscosity is therefore indicative of the amount of amylose leached out of the granules.

The properties of wheat starch are useful in a large number of applications and also non-food (paper, textiles, adhesives etc.) applications. However, for many applications, properties are not optimum and various chemical and physical modifications well known in the art are undertaken in order to improve useful properties. Two types of property manipulation which would be of use are: the controlled alteration of gelatinisation and pasting temperatures; and starches which suffer less granular fragmentation during pasting than conventional starches.

Currently the only ways of manipulating the gelatinisation and pasting temperatures of starch are by the inclusion of additives such as sugars, polyhydroxy compounds of salts or by extensive physical or chemical pre-treatments. The reduction of granule fragmentation during pasting can be achieved either by extensive physical pre-treatments

or by chemical cross-linking. Such processes are inconvenient and inefficient. It is therefore desirable to obtain plants which produce starch which intrinsically possesses such advantageous properties.

Starch consists of two main glucose polysaccharides: amylose and amylopectin. Amylose is a generally linear polymer comprising α-1,4 linked glucose units, while amylopectin is a highly branched polymer consisting of an α-1,4 linked glucan backbone with α-1,6 linked glucan branches. In wheat endosperm amylopectin constitutes approximately 70% of the total starch content, with the balance being amylose. Amylopectin is synthesised through the concerted action of several enzymes, including soluble starch synthase(s) (SSS), starch branching enzyme(s) (SBE), starch de-branching enzyme(s) (DBE). The physical properties of starch are strongly affected by the relative abundance of amylose and amylopectin, therefore SSSs, SBEs and DBEs play a key role in determining both starch quantity and quality. As such, one approach to manipulating starch structure would be to modify the expression of the enzymes involved in starch biosynthesis in the endosperm using a transgenic approach.

SBE catalyses the formation of the α-1,6 linkages, creating branch points in the growing starch molecule, via hydrolysis of an α-1,4 linkage followed by reattachment of the released α-1,4-glucan chain to the same or another glucosyl chain. This reaction also provides a new non-reducing end for further elongation of the original α-1,4-glucan chain.

Multiple isoforms of starch branching enzyme have been described, biochemically, from a number of species including maize (Boyer and Preiss, 1978), rice (Nakamura *et al.*, 1992), pea (Smith, 1988), potato (Khoshnoodi *et al.*, 1993) and wheat (Morell *et al.*, 1997). More recently, genomic and cDNA sequences for SBE have been characterised from several species including maize (Baba *et al.*, 1991; Fisher *et al.*, 1993; Gao *et al.* 1997) pea (Burton *et al.*, 1995), potato (Kossmann *et al.*, 1991), rice (Nakamura and Yamanouchi, 1992; Mizuno *et al.*, 1993), *Arabidopsis* (Fisher *et al.*, 1996), cassava (Salehuzzaman *et al.*, 1992), and wheat (Rapellin *et al.*, 1997, Nair *et al.*, 1997, Rahman *et al.*, 1997). Sequence alignment of these SBEs revealed a high degree of sequence conservation at the amino acid level and that the SBEs may be grouped into two distinct

families, generally known as SBEI and SBEII. Further, analysis indicates that within a species there is generally of the order of 50% homology between the two families, SBEI and SBEII, while there is often greater homology within the two families between species.

Maize is unusual in that the maize SBEII family is thought to comprise two different members, known as SBEIIa and SBEIIb. There has been controversy over whether the SBEIIa and IIb enzymes are in fact a) encoded by genes at two different loci, and b) whether the genes represent different alleles at a single locus. Fisher et al (1996) and Gao et al (1997) have provided evidence that SBEIIa and SBEIIb are encoded by independent genes. However, there is no conclusive evidence that both isoforms exist together in any one maize genotype. The DNA clones for the two published gene sequences were purified from different genotypes of maize and it is thus possible that they represent different alleles of a single locus. In summary, in maize, three distinct SBE genes have been characterised to date (Baba *et al.*, 1991; Fisher *et al.*, 1993; Gao *et al.*, 1997). SBEI is distinct from SBEIIa and SBEIIb in amino acid composition, substrate specificity, kinetic properties, and immunological reactivities, whereas SBEIIa and SBEIIb are similar in these respects (Guan and Preiss, 1993; Preiss 1991; Takeda *et al.*, 1993). At the amino acid level the sequence exhibits approximately 50% homology with the SBEIIa and SBEIIb sequences, whereas SBEIIa and SBEIIb exhibit approximately 80% homology to each other.

Prior to the present invention, maize was unique in having SBEIIa- and SBEIIb-type enzymes. Although *Arabidopsis* has two SBEII family members, the sub-division in *Arabidopsis* does not appear to conform to that seen in maize: the *Arabidopsis* sub-family members do not obviously fall into the IIa and IIb categories as do the maize sequences. Both of the *Arabidopsis* SBEII genes have similar levels of homology to both the maize SBEII genes, SBEIIa and SBEIIb, but the similarities are not sufficient to be able to place the *Arabidopsis* genes into the same SBEIIa and SBEIIb categories as for maize. Indeed, the data, if anything, suggests that the *Arabidopsis* SBEII genes do not fall into the maize IIa and IIb categories. For barley, two forms of SBEII had been partly characterised. Although these have been called SBEIIa and SBEIIb, only a very limited amount of sequence information had been published (Sun *et al*, 1995) and it was not possible to infer

or conclude that these forms correspond to the IIa and IIb categories of maize. In fact, based on the available barley sequence information both of the barley SBEII sequences (SBEIIa and SBEIIb) would appear to show greater homology to maize SBEIIa than to maize SBEIIb.

For all other plant species for which SBEII sequences have been identified and published, including potato, pea, rice, cassava, wheat and barley, no sub-division of the SBEII family comparable to the SBEIIa and SBEIIb division of maize has been made.

Studies of purified SBEI and SBEII demonstrate that these isoforms differ in their specificity for a substrate with respect to both chain length and degree of branching. In maize, SBEI and SBEII show distinct branching activities *in vitro*, with SBEI showing a higher rate of branching of an amylose substrate when compared to SBEII whereas both SBEIIa and IIb show higher rates of branching than SBEI when acting upon an amylopectin substrate (Guan and Preiss, 1993). Furthermore, maize SBEI preferentially transfers longer glucan chains (average chain length = 24) than SBEII (average chain length = 21(IIa) and 22(IIb)) (Takeda *et al.*, 1993). A similar observation has been reported for SBEI and SBEII isoforms from wheat and pea (Morell *et al.*, 1997; Smith, 1988). Mutational studies in maize, rice and pea demonstrate that high amylose mutants in each case are deficient in the branching enzyme activity analogous to maize SBEII (Martin and Smith, 1995; Morell *et al.*, 1995). However, the linkage between the biochemical observations and the genetic evidence suggesting the differences in the roles remains unclear.

The present invention is based on the unexpected discovery of a novel class of SBEII genes in wheat, referred to herein as SBEII-1. The novel SBEII-1 gene sequence has strong homology with the maize SBEIIb gene. The wheat SBEII-1 genes are thought to be functionally equivalent to the maize SBEIIb gene, and on this basis it is believed that manipulation of the wheat SBEII-1 gene is likely to influence starch properties including starch gelatinisation temperature, in a manner analogous to manipulation of the maize SBEIIb gene as described in WO 97/22703.

In summary, although two different SBEII gene sequences are known from maize, Arabidopsis and barley, as discussed above, prior to the present invention there was no reason to expect that wheat would show a similar sub-division of SBEII genes as is seen for maize. The two Arabidopsis SBEII genes show a different sub-division, and prior to the present invention there was insufficient evidence to determine whether the two barley SBEII sequences belonged to the maize-type sub-division. That is, prior to the present invention there was no reason to expect that wheat would have two similar SBEII members comparable to those of maize. Subsequent to the present invention Sun et al (1998) have presented data which indicates that the barley sequences do indeed sub-divide in a similar manner to the maize SBEIIa and IIb sequences and the wheat SBEII-2 and SBEII-1 sequences discussed in this document.

The present inventors have used the high degree of sequence conservation between several SBE gene sequences to design oligonucleotide primers to motifs which are specific to either SBEI or SBEII families and have used these primers to amplify cDNA sequences from developing endosperm of wheat.

When this work was started, a single partial length wheat SBE cDNA clone had been reported (Mousley, 1994). Multiple sequence alignment of this wheat SBE sequence with other published SBE sequences from a number of plant species revealed a number of motifs which were highly conserved. Oligonucleotide primers designed to be complementary to these motifs were used to clone 3' partial length cDNA clones of wheat SBEII. Alignment of the cDNA clone sequences indicated that the clones could be divided into two classes, which the inventors have designated SBEII-1 and SBEII-2, which showed greater than 90% similarity to members within a class but only 60% similarity between classes. Significantly, comparison between representative sequences from each class with previously identified wheat SBEII clones, pWBE6 (Mousley, 1994) and SBEII (Nair *et al.*, 1997), showed that each appear to be homologues of the SBEII-2 class. The cloning of a wheat SBEII-1 cDNA is novel.

Summary of the Invention

In one aspect the invention provides a nucleotide sequence encoding substantially the amino acid sequence shown in Figure 10 (SEQ ID No: 2) or a functional equivalent of said nucleotide sequence.

The term functional equivalent is used in this context to encompass those sequences which differ in their nucleotide composition to that shown in Figure 10 (SEQ ID No: 1) but which, by virtue of the degeneracy of the genetic code, encode polypeptides having identical or substantially identical amino acid sequences. It is intended that the term should generally apply to sequences which are sufficiently homologous to the sequence of the invention that they can hybridise to the complement thereof under stringent hybridisation conditions (eg as described by Sambrook et al 1989, ie washing with 0.1xSSC, 0.5% SDS at 68°C); such equivalents will preferably possess at least 86%, more preferably at least 90%, and most preferably at least 95%, sequence homology (ie sequence similarity) with the sequence of the invention. Sequence homology is suitably determined using the 'MEGALIGN' program of the software package DNAStar (MEGALIGN and DNAStar are Trade Marks). It will be apparent to those skilled in the art that the nucleotide sequence of the invention may also find useful application when present as an "antisense" sequence. Accordingly, functionally equivalent sequences will also include those sequences which can hybridise, under stringent hybridisation conditions, to the sequence of the invention (rather than the complement thereof). Such "antisense" equivalents will preferably possess at least 86%, more preferably at least 90%, and most preferably 95% sequence homology with the complement of the sequence of the invention.

In another aspect, the invention provides a nucleotide sequence comprising substantially the sequence of B2 shown in Figure 3 (SEQ ID No: 3), or a functional equivalent thereof.

In a further aspect, the invention provides a nucleotide sequence comprising substantially the sequence of B4 shown in Figure 3 (SEQ ID No: 4), or a functional equivalent thereof.

Another aspect of the invention provides a nucleotide sequence comprising substantially

the sequence of B10 shown in Figure 3 (SEQ ID No: 5), or a functional equivalent thereof.

Yet a further aspect of the invention provides a nucleotide sequence comprising substantially the sequence of B1 shown in Figure 3 (SEQ ID No: 6), or a functional equivalent thereof.

In another aspect the invention provides a nucleotide sequence encoding substantially the amino acid sequence of B6 shown in Figure 4 (SEQ ID No: 7), or a functional equivalent thereof.

The term functional equivalent in this context has the same general meaning as discussed above, although equivalents for B2, B4, B10 and B6 will preferably possess at least 90%, more preferably at least 95%, sequence homology with the relevant sequence of the invention, while equivalents for B1 will preferably possess at least 97% sequence homology with the sequence of the invention.

The sequences of the invention are part of novel wheat SBEII genes, with B1 being a novel subclass of the known class of SBEII genes, referred to herein as SBEII-2, with the novel subclass being called SBEII-2B. The remaining sequences are all of a completely new class of wheat SBEII genes, referred to herein as SBEII-1. The sequences have been found to fall into 3 sub-classes, to be discussed below.

The novel wheat SBEII-1 genes that are the subject of this invention have strong sequence homology with the maize SBEIIb gene. The wheat SBEII-1 genes are thought to have similar functional properties to the maize SBEIIb gene. On this basis it is expected that by genetic manipulation of the wheat SBEII-1 gene it will be possible to influence properties of starch produced by a plant, including the gelatinisation temperature and rheological properties of starch, in a manner analogous to manipulation of the maize SBEIIb gene described in WO 97/22703. The content of WO 97/22703 is incorporated herein by reference.

The present invention also includes within its scope a portion of any of the above sequences, comprising at least 500 base pairs and having at least 90% sequence homology to the corresponding portion of the sequence from which it is derived.

Although the coding sequences of the novel wheat SBEII-1 genes have strong sequence homology with the maize SBEIIb gene, there is much greater divergence in the 3' untranslated parts of the sequences, with a maximum of 31.8% homology between the 3' untranslated sequences of wheat SBEII-1 and maize SBEIIb as is apparent from Figure 8.

In another aspect the invention thus provides a nucleotide sequence comprising substantially the sequence shown in Figure 5 (SEQ ID No: 8), Figure 6 (SEQ ID No: 9) or Figure 7 (SEQ ID No: 10), or a functional equivalent thereof.

The term functional equivalent in this context has the same general meaning as discussed above, but with equivalents preferably at least 32%, more preferably at least 40%, 50%, 60%, 70%, 80% or 90% sequence homology with the sequence of the relevant Figure.

It is thought such 3' untranslated sequences may be useful, both in sense and antisense function, in manipulation of starch properties by affecting SBE expression in plants, as will be discussed below.

The sequence may include further nucleotides at the 5' or 3' end. For example, for ease of expression, the sequence desirably also comprises an in-frame ATG start code, and may also encode a leader sequence.

The invention also covers a nucleic acid construct comprising a nucleotide sequence or portion thereof in accordance with the invention conveniently operably linked, in sense or antisense orientation, to a promoter sequence.

Also included within the scope of the invention is amino acid sequence encoded by any of the nucleotide sequences of the invention.

The invention also provides vectors, particularly expression vectors, comprising the nucleotide sequence of the invention. The vector will typically comprise a promoter and one or more regulatory signals of the type well known to those skilled in the art. The invention also includes provision of cells transformed (which term encompasses transduction and transfection) with a vector comprising the nucleotide sequence of the invention.

Nucleotide sequences in accordance with the invention may be introduced into plants, particularly but not exclusively wheat plants, and it is expected that this can be used to affect expression of SBE in the plant and hence affect the properties of starch produced by the plant. In particular, use of sequences in antisense orientation is expected to reduce or suppress enzyme expression. Additionally, it has recently been demonstrated in other experimental systems that "sense suppression" can also occur (i.e. expression of an introduced sequence operably linked in the sense orientation can interfere, by some unknown mechanism, with the expression of the native gene), as described by Matzke & Matzke 1995. Any one of the methods mentioned by Matzke & Matzke could, in theory, be used to affect the expression in a host of a homologous SBE gene.

It is believed that antisense methods are mainly operable by the production of antisense mRNA which hybridises to the sense mRNA, preventing its translation into functional polypeptide, possibly by causing the hybrid RNA to be degraded (e.g. Sheehy *et al.*, 1988; Van der Krol *et al.*,). Sense suppression also requires homology between the introduced sequence and the target gene, but the exact mechanism is unclear. It is apparent however that, in relation to both antisense and sense suppression, neither a full length nucleotide sequence, nor a "native" sequence is essential. Preferably the "effective portion" used in the method will comprise at least one third of the full length sequence, but by simply trial and error other fragments (smaller or larger) may be found which are functional in altering the characteristics of the plant.

Thus, in a further aspect the invention provides a method of altering the characteristics of a plant, comprising introducing into the plant an effective portion of the sequence of the invention operably linked to a suitable promoter active in the plant so as to affect

expression of a gene present in the plant. Conveniently the sequence will be linked in the antisense orientation to the promoter. Preferably the plant is a wheat plant. Conveniently, the characteristic altered relates to the starch content and/or starch composition of the plant (i.e. amount and/or type of starch present in the plant). Preferably the method of altering the characteristics of the plant will also comprise the introduction of one or more further sequences, in addition to an effective portion of the sequence of the invention. The introduced sequence of the invention and the one or more further sequences (which may be sense or antisense sequences) may be operably linked to a single promoter (which would ensure both sequences were transcribed at essentially the same time), or may be operably linked to separate promoters (which may be necessary for optimal expression). Where separate promoters are employed they may be identical to each other or different. Suitable promoters are well known to those skilled in the art and include both constitutive and inducible types. Examples include the CaMV 35S promoter (e.g. single or tandem repeat) and the ubiquitin promoter. Advantageously the promoter will be tissue-specific. Desirably the promoter will cause expression of the operably linked sequence at substantial levels only in the tissue of the plant where starch synthesis and/or starch storage mainly occurs.

The sequence of the invention, and the one or more further sequences if desired, can be introduced into the plant by any one of a number of well-known techniques (e.g. *Agrobacterium*-mediated transformation, or by "biolistic" methods). The sequences are likely to be most effective in affecting SBE activity in wheat plants, but theoretically could be introduced into any plant. Desirable examples include pea, tomato, maize, rice, barley, sweet potato and cassava plants. Preferably the plant will comprise a natural gene encoding an SBE molecule which exhibits reasonable homology with the introduced nucleic acid sequence of the invention.

In another aspect, the invention provides a plant cell, or a plant or the progeny thereof, which has been altered by the method defined above. The progeny of the altered plant may be obtained, for example, by vegetative propagation, or by crossing the altered plant and reserving the seed so obtained. The invention also covers parts of the altered plant, such as storage organs. Conveniently, for example, the invention covers grain comprising

altered starch, said grain being obtained from an altered plant or the progeny thereof. Grain obtained from altered plants (or the progeny thereof) will be particularly useful materials in certain industrial applications and for the preparation and/or processing of foodstuffs and may be used, for example, in bakery products.

In particular relation to wheat plants, the invention provides a wheat plant or part thereof which, in its wild type possesses an effective SBEII-1 gene, but which plant has been altered such that there is either reduced, increased or no effective expression of an SBEII-1 polypeptide within the cells of at least part of the plant. The plant may have been altered by the method defined above, or may have been selected by conventional breeding to be deleted for the SBEII-1 gene, the presence or absence of which can be readily determined by screening samples of the plants with a nucleic acid probe or antibody specific for the wheat gene or gene product respectively.

The invention also provides starch extracted from a plant altered by the method defined above, or from the progeny of such a plant, the starch having altered properties compared to starch extracted from equivalent, but unaltered, plants. The invention further provides a method of making altered starch, comprising altering a plant by the method defined above and extracting therefrom starch having altered properties compared to starch extracted from equivalent, but unaltered, plants. It is believed that use of nucleotide sequences in accordance with the invention will enable the production of starches, particularly wheat starches, having a wide variety of novel properties. For example, it may be anticipated that plants altered to give a reduction in SBEII activity will give rise to a starch with a relatively higher proportion of amylose and a lower proportion of amylopectin compared with that from unaltered plants.

In particular the invention provides the following: a plant (especially a wheat plant) altered by the method defined above, containing starch which, when extracted from the plant, has an elevated gelatinisation onset and/or peak temperature as measured by DSC, compared to starch extracted from a similar, but unaltered, plant; a plant (especially a wheat plant) altered by the method defined above, containing starch which, when extracted from the plant, has a elevated gelatinisation onset temperature (conveniently elevated by at least

3°C, possibly by at least 7°C, by at least 12°C or possibly even by 15 to 25°C) as measured by DSC compared to starch extracted from a similar, but unaltered plant; a plant (especially a wheat plant) altered by the method defined above, particularly to reduce expression of SBEII-1 polypeptide, containing starch which, when extracted from a plant, has a higher amylose:amylopectin ratio compared to starch extracted from a similar, but unaltered plant.

The present invention particularly covers starch extracted from a plant altered by the method of the invention, particularly starch having an increased gelatinisation temperature. Such starch is useful, eg in bakery products, having particular benefits in certain situations, and the invention also covers products, particularly bakery products, made from such starch. The invention also covers starch extracted from a plant altered by the method of the invention and having an increased amylose:amylopectin ratio.

The invention will be further described, by way of illustration, in the following Examples and with reference to the accompanying drawings, in which:

Figure 1 is a graph of viscosity versus time, showing a viscoamylgraph profile for wheat starch during and after cooking;

Figure 2 shows alignment amino acid sequence data of C terminal portions of various known starch branching enzymes (SEQ ID Nos: 12 to 25), obtained from the European Molecular Biology Laboratory (EMBL) database, and for a novel wheat SBEII-1 sequence of the invention (OsbeII-1ALL) (SEQ ID No: 11) from clone 5A1, with consensus residues highlighted;

Figure 2a is a residue weight table showing the percent similarity and percent divergence of the sequences shown in Figure 2;

Figure 3 shows aligned DNA sequence data for various recombinant clones (B2, B4, B10, A2, B1, B11) (SEQ ID Nos: 3, 4, 5, 26, 6, 27 respectively) containing wheat starch branching enzyme genes, representing two SBE classes, SBEII-1 and SBEII-2, each of

which includes three subclasses A, B and C, with residues differing from the consensus (majority) (SEQ ID No: 53) highlighted;

Figure 3a is a residue weight table showing the percent similarity and percent divergence of the sequences shown in Figure 3;

Figure 4 is an alignment of predicted amino acid sequences for clones B6 (wheat SBEII-1) (SEQ ID No: 7) and B11 (wheat SBEII-2) (SEQ ID No: 28) against the corresponding regions of the maize SBEIIa (SEQ ID No: 29) and SBEIIb (SEQ ID No: 30) amino acid sequences, with residues differing from those of maize SBEIIb highlighted;

Figure 4a is a residue weight table showing the percent similarity and percent divergence of the sequences shown in Figure 4;

Figure 5 shows the 3' untranslated DNA sequence of clone B2 (SEQ ID No: 8) (wheat SBEII-1, sub-class A);

Figure 6 shows the 3' untranslated DNA sequence of clone B10 (SEQ ID No: 9) (wheat SBEII-1, sub-class B);

Figure 7 shows the 3' untranslated DNA sequence of clone B4 (SEQ ID No: 10) (wheat SBEII-1, sub-class C);

Figure 8 shows aligned DNA sequence data for the 3' untranslated region of clones B10 (SEQ ID No: 9), B2 (SEQ ID No: 8) and B4 (SEQ ID No: 10) and maize SBEIIb (ZMSBE2b) (SEQ ID No: 31), with residues differing from those of the B10 sequence highlighted;

Figure 8a is a residue weight table showing the percent similarity and percent divergence of the sequences shown in Figure 8;

Figures 9a and 9b show hybridisation of clone B1 (SBEII-2) and clone B2 (SBEII-1),

respectively, to HindIII-digested genomic DNA of Chinese Spring wheat nullisomic-tetrasomic lines;

Figure 10 shows the DNA (SEQ ID No: 1) and predicted amino acid sequence (SEQ ID No: 2) of part of SBEII-1 clone 5A1;

Figure 11 shows aligned amino acid sequence data for the wheat SBEII-1 sequence of the invention, from clone 5AI (OsbeII-1ALL) (SEQ ID No: 11), wheat SBEI-D2 (SEQ ID No: 32) of Rahman *et al* 1997 (TASBEID2), wheat SBE1 of Rapellin *et al* 1997 (SEQ ID No: 33) (TASBEI) and wheat SBEII-2 of Nair *et al* 1997 (SEQ ID No: 34) (wheat SBEII-2), with residues exactly matching the consensus (majority) (SEQ ID No: 54) highlighted;

Figure 11a is a residue weight table showing the percent similarity and percent divergence of the sequences shown in Figure 11;

Figure 12 illustrates northern blotting of wheat grains harvested at various different intervals after anthesis and probed with SBEII-1 and SBEII-2 fragments;

Figure 13 is a restriction map of plasmid pWxGS+;

Figure 13a shows the sequence (SEQ ID No: 55) of the promoter (HindIII-BamH1 fragment) in pWxGS+;

Figure 14 is a restriction map of plasmid pSRWXGUS1;

Figure 15 is a restriction map of plasmid pVTWXGUS2;

Figure 16 is a restriction map of plasmid pPBI-97-2;

Figure 17 is a restriction map of plasmid pSR97-26A-;

Figure 18 is a restriction map of plasmid pSR97-29A-;

Figure 19 is a restriction map of plasmid pSR97-50A-;

Figure 20 is a restriction map of plasmid pSR97-53A-;

Figure 21 is a restriction map of plasmid p97-2C;

Figure 22 is a restriction map of plasmid p97-2CWT1;

Figure 23 is a restriction map of plasmid pSC98-1;

Figure 24 is a restriction map of plasmid pSC98-2;

Figure 25 is a restriction map of plasmid pUNI;

Figure 26 shows the DNA sequence of the NptII Sac1 fragment of pUNI (SEQ ID No: 35); and

Figure 27 is a restriction map of plasmid pUSN99-1;

Figure 28 is a restriction map of plasmid pUSN99-2;

Figure 29 is a partial restriction map of the predicted sequence (SEQ ID No: 52) of a cloned fragment of p97-U3;

Figure 30 is a restriction map of plasmid pPBI96-36;

Figure 31 is a restriction map of plasmid p97-dUG1;

Figure 32 is a restriction map of plasmid p97-2BdUN1;

Figure 33 is a schematic illustration of a particle bombardment chamber (not to scale);

Figure 34 shows histochemical localisation of Ubi-GUS expression in seed (panel A), stem (panel B), floral (panel C) and leaf tissues (panel D) of wheat transformed with plasmid pAHC25;

Figure 35 is a Southern blot of 26 progeny plants of transformant BW119 which had been transformed with pAHC25.

Figure 36 shows histochemical localisation of waxy-GUS expression in endosperm tissue of two independent transgenic wheat lines (in panels A and B) transformed with the plasmid pWxGS+; and

Figure 37 is a Southern blot of genomic DNA of putative primary transformants digested with Sac1 and probed with the 1kb Sac1 SBEII-1 probe.

Examples

Amplification and characterisation of two classes of SBEII cDNA clones

A PCR based cloning strategy was devised for isolating starch branching enzymes from wheat using conserved domains within the known cloned gene sequences. Starch branching enzymes have been cloned from a number of plant species and Figure 2 shows amino acid sequence data, obtained from the European Molecular Biology Laboratory (EMBL) nucleotide database for various known starch branching enzymes as follows:-

Wheat SBEII-2 for *Triticum aestivum* (SEQ ID No: 12)
ZM SBE2a (maize) for *Zea mays* (SEQ ID No: 13)
ZM SBE2b (maize) for *Zea mays* (SEQ ID No: 14)
Barley SBEIIa (SEQ ID No: 15)
Barley SBEIIb (SEQ ID No: 16)
RICBCE3 (rice SBEII type enzyme) for *Oryza sativa* (SEQ ID No: 17)

RICESBE-1/97 (as above, including transit peptide sequence) (SEQ ID No: 18)
PSSBEIGEN (pea SBEI, which is in fact an SBEII- type sequence) for *Pisum sativum* (SEQ ID No: 19)
STSBE (potato SBEI type) for *Solanum tuberosum* (SEQ ID No: 20)
TASBEI (wheat SBEI-2) for *Triticum aestivum* (SEQ ID No: 21)
TASBEI D2 (SEQ ID No: 22)
ZMSBEI (maize SBEI) for *Zea mays* (SEQ ID No: 23)
RICBEI (rice SBEI) for *Oryza sativa* (SEQ ID No: 24)
PSSBEIIGN (pea SBEII, which is in fact an SBEI-type sequence) for *Pisum sativum* (SEQ ID No: 25)

Figure 2 also shows sequence information for a novel wheat SBEII-1 sequence of the invention, identified as OsbeII-1ALL (SEQ ID No: 11).

The alignment report of Figure 2, and also Figures 3, 4, 8 and 11, was prepared using Clustal method, with PAM 250 residue weight table for amino acid sequences and weighted residue weight table for DNA sequences. Sequence pair distances expressed as % similarity shown in Figures 2A and 3A, 4A, 8A and 11A are determined using a 'MEGALIGN' program of DNAStar software, and correspond to sequence homology percentages as specified above.

Alignment of the sequences shown in Figure 2 reveals several domains which are highly conserved. One such domain, MDKDMYD (SEQ ID No: 36), was almost completely conserved and it was assumed that this domain would also be present in wheat starch branching enzyme genes. This motif was chosen as a target for an oligonucleotide sense primer (SBEA). 3'RACE PCR was carried out on endosperm first strand cDNA using the primers Ro and SBE A.

Two populations of PCR products of approximately 1kb and 1.2Kb were cloned into the plasmid vector pT7Blue (Novagen). Plasmid DNA from 36 putative recombinant clones was purified and the insert size estimated by restriction analysis. Fifteen clones harbouring inserts of between approximately 1Kb and 1.2Kb were selected for sequencing.

Alignment of the sequence data obtained, using the MEGALIGN program of DNAStar, indicated that the 15 selected clones could be divided on the basis of degrees of homology into two different classes, which we have designated SBEII-1 and SBEII-2. Furthermore, both the SBEII-1 and SBEII-2 classes may each be further subdivided into three sub-classes, based on sequence differences (Table 1). It is thought the sub-division into three sub-classes probably arises because wheat comprises three homoeologous genomes.

Table 1

Class	Sub-Class	Clone Number
SBEII-1	A	B2, B5, B6, B7, B12
SBEII-1	B	B10
SBEII-1	C	A1, A13, B4
SBEII-2	A	B11
SBEII-2	B	B1, B9
SBEII-2	C	A2, C5

Comparison between sequences within either of the SBEII-1 or SBEII-2 classes showed between 90 and 96.8% similarity. In contrast, sequence similarity between representatives of SBEII-1 and SBEII-2 classes only display between 58.8 and 60.0% homology in the region of comparison (Figures 3 and 3a).

Furthermore, we have compared representative sequences from each SBEII-1 and SBEII-2 class with the previously reported wheat SBEII clones, pWBE6 (Mousley, 1994) and the very recently published SBEII (Nair *et al.*, 1997). The results showed that each of the previously isolated SBEII clones are highly homologous (>90%) to our SBEII-2 class (data not shown). Significantly, neither of the previously reported wheat sequences showed high homology to our SBEII-1 sequence. The isolation and characterisation of three forms of SBEII-1 (SBEII-1, sub-classes A, B & C) is novel. The SBEII-2 sub-class B is also novel, sub-classes A and C corresponding to the sequences previously disclosed by Mousley (1994) and Nair *et al* (1997) respectively.

Alignment of the predicted amino acid sequences from representative clones, B6 and B11 of the wheat SBEII-1 and SBEII-2 sequences (respectively) against the corresponding regions of the maize SBEIIa and SBEIIb amino acid sequences (Figure 4 and 4a) indicate that the wheat SBEII-1 sequence (clone B6) is more similar to the maize SBEIIb sequence (88.7% similarity) than to the wheat SBEII-2 sequence and the maize SBEIIa sequence (82.2% & 82.6% similarity respectively) and similarly that the wheat SBEII-2 sequence is more similar to the maize SBEIIa sequence (86.9% similarity) than to the wheat SBEII-1 and maize SBEIIb sequences (82.2% and 81.7% similarity respectively). We thus hypothesise that the wheat SBEII-1 is phylogenetically more related to the maize SBEIIb and that the wheat SBEII-2 is phylogenetically related to the maize SBEIIa sequences and that the corresponding wheat and maize sequences are likely to exhibit similar functional properties.

While the coding sequences of clones B2, B10 and B4 have strong sequence homology to the maize SBEIIb gene, there is much greater divergence in the 3' untranslated parts of the sequences. Figure 5, 6 and 7 show the 3' untranslated sequences of clones B2, B10 and B4, respectively, and Figure 8 compares these sequences with the corresponding sequence of maize SBEIIb.

Considering matters in more detail, experimental details were as follows.

Plant material

Triticum aestivum cultivar Rialto was grown in a glass house under supplementary lighting and temperature control to maintain a 16 hour day-length at 18+/-1°C.

Recombinant DNA manipulations and sequencing

Standard procedures were performed essentially according to Sambrook *et al.*, (1989). DNA sequencing was performed on an ABI automated sequencer and sequences analysed using DNASTAR software for Macintosh.

RNA isolation for cDNA cloning

RNA was extracted from *Triticum aestivum* cultivar Rialto endosperm, using a Purescript RNA isolation kit (Flowgen) essentially according to the manufacturers recommendations. Briefly, endosperm tissue was frozen in liquid nitrogen and ground, for 2 min, to a fine powder using a dismembrenator (Braun Biotech International). The ground tissue was stored in liquid nitrogen prior to extraction. Approx. 100mg of ground tissue was transferred to a 1.5ml microcentrifuge tube and 1.2ml of 'Lysis buffer' was added to the tissue before mixing by inversion and placing on ice for 10 minutes. Protein and DNA were precipitated from the cell lysate by adding 0.4ml of 'Protein-DNA Precipitation Solution' and mixing by inversion before centrifuging at 13,000 x *g* at 4°C for 20 minutes. The supernatant was divided between two fresh 1.5ml tubes each containing 600μl of *iso*-propanol. The RNA precipitate was pelleted by centrifugation at 13,000 x *g* at 4°C for 10 minutes, the supernatant was discarded and the pellets washed with 70% ethanol by inverting the tube several times. The ethanol was discarded and the pellet air dried for 15-20 minutes before the RNA was resuspended in 7.5ml of 'RNA Hydration Solution'.

Preparation of wheat endosperm cDNA pool

Wheat endosperm cDNA pool was prepared from total RNA, extracted as described above, using Superscript™ reverse transcriptase (Life Technologies) essentially according to manufacturers instructions. Briefly, five microgrammes of RNA, 10pMol RoRidT17 [AAGGATCCGTCGACATCGATAATACGACTCACTATAGGGA(T17)] (SEQ ID No: 37) and sterile distilled water to a reaction volume of 12μl, in a 500μl microcentrifuge tube, was heated to 70°C for 10 minutes before being quick chilled on ice. The contents of the tube were collected by brief centrifugation before adding 4μl 5x First Strand Buffer, 2μl 0.1M DTT and 1μl 10mM dNTPs and, after mixing, incubating at 42°C for 2 min. 1μl of Superscript™ was added and, after mixing, incubation continued for 1 hour. The reaction was inactivated by heating to 70°C for 15 min. 150μl of $T_{10}E_1$ was added to the reaction mix and the resulting cDNA pool was used as a template for amplification in PCR.

PCR amplification of SBEII sequences from endosperm cDNA pool

SBEII sequences were amplified from the endosperm cDNA pool using primers Ro [AAGGATCCGTCGACATC] (SEQ ID No: 38), which is complementary to the Ro region of the RoRidT17 primer used to synthesise the cDNA pool, and the SBEII specific primer, SBEA [ATGGACAAGGATATGTATGA] (SEQ ID No: 39). SBEA was designed to be homologous to the MDKDMYD (SEQ ID No: 36) motif which is situated approx. 1kb from the 3'end of the mature peptide coding sequence. PCR was carried out in a 50μl reaction, comprising 5μl of the cDNA pool, 25pmol Ro, 50pmol SBEA, 5μl 5x Taq buffer, 4μl 25mM Mg^{2+}, 0.5μl 20mM dNTPs, and 1.25u Taq polymerase. All of the reaction components were mixed, except for the Taq polymerase, before being pre-heated to 94°C for 7 min and then cooled to 75°C for 5 min. Whilst the reaction mixtures were held at 75°C the Taq polymerase was added and, after mixing well, the reactions were thermocycled at (94°C-30sec, 50°C-30sec, 72°C-1min) x 30 cycles, followed by a final 10 min extension step at 72°C.

PCR products were purified by phenol/chloroform and chloroform extraction before ligation with pT7 Blue (Novagen) according to manufacturers recommendations. Putative SBE clones were initially characterised by standard plasmid DNA purification methods and restriction digestion. Representative clones harbouring a range of different sized inserts were selected for sequencing.

Chromosomal location of SBE genes in wheat

The Chinese Spring wheat nullisomic-tetrasomic lines as described in Sears (1966) were used for assignment of the SBE sequences chromosome locations. Ditelosomic lines (Sears, 1966) were used to determine the chromosome arm location. The Betzes barley ditelosomic addition lines in wheat are described in Islam (1983).

The chromosomal location of the two families of SBEII sequences (SBEII-1, SBEII-2) was determined by probing wheat nulli-tetra and ditelosomic stock lines with gel-purified inserts of the various clones. Figure 9a shows the hybridisation obtained with an SBEII-2

(clone B1) probe on HindIII digested DNA. The euploid Chinese Spring gives 3 bands, one of which is missing in turn in the lines nullisomic for chromosomes 2A, 2B and 2D. The same blot was re-probed with a SBEII-1 specific probe (clone B2). This yields an entirely different hybridisation profile (Figure 9b), demonstrating the specificity of the probe used. Again bands are missing in each of the lines nullisomic for 2A, 2B and 2D. the same banding pattern was observed using the SBEII-1 clones B2 and B4. Thus the SBEII sub-family 1 and 2 gene sequences lie on the wheat group 2 set of homeologous chromosomes.

Ditelosomic addition lines were used to identify the arm location of these genes (data not shown). This revealed that the SBEII-1 and SBEII-2 sequences are both located on the long arms of the homeologous group 2 chromosomes of wheat.

Barley addition lines were used to determine whether homologous sequences are present in barley. These showed that sequences homologous to the wheat SBEII-1 and SBEII-2 sequences are located on the long arms of barley chromosome 2H.

RNA Isolation and Northern Blotting

Wheat grains were harvested at appropriate intervals and frozen in liquid Nitrogen before grinding to a fine powder using either a Braun Mikrodismembrator™ or a pestle and mortar. Total RNA was isolated using the RNAqueous™ (Ambion Inc) Kit according to the manufacturers instructions, or with the following method. Frozen powdered grain was mixed with a 10X volume of 0.2M Tris-HCl pH9, 0.4M NaCl, 25mM EDTA, 1% SDS, 1% PVPP, 0.25% Antifoam A, and 0.1M DTT. This mixture was extracted twice with an equal volume of phenol/chloroform/isoamyl alcohol (25:24:1), the nucleic acids precipitated from the aqueous phase by the addition of 0.8 volumes of isopropanol, and the resulting pellet dissolved in H_2O. The RNA was then selectively precipitated by the addition of 1 volume of 4M LiCl, incubated at 4°C overnight, and the resulting pellet dissolved in sterile distilled H_2O. 15 μg of total RNA was electrophoresed on a 1% agarose, 2.21M Formaldehyde, 40mM MOPS pH7.0, 10mM sodium acetate, 1mM EDTA gel, in a 40mM MOPS pH7, 10mM sodium acetate, 1mM EDTA running buffer at 1

V/cm overnight. Gels were placed in a 50ng/ml solution of Ethidium Bromide in water for 30 minutes, de-stained in water for 2 hours, and visualised and photographs under UV light. The gels were then washed briefly in sterile distilled H_2O, then blotted onto HyBond N^{+TM} (Amersham International), according to standard protocols (Sambrook et al, 1989) overnight. Blots were then dismantled and air-dried before UV fixing at 312nm for 2 minutes.

Probe Isolation and Purification

5-10 μg of the plasmids pUN1 and pSR98-29 were digested with Sst1 (Life Technologies Ltd) according to the manufacturers instructions, to release fragments of approximately 0.8kb (NptII) and 1kb (SBEII-1) respectively. 5-10μg of the plasmid pVT96-54 was digested with BamH1 to release a SBEII-2 fragment of approximately 1.2kb. Digests were electrophoresed on 1% low melting point agarose gels. The gene specific fragments were excised and the DNA purified using a Wizard™ Gel Purification Kit (Promega).

Probe Labelling and Hybridization

25ng of the appropriate probe (Maize Waxy promoter, NptII, Wheat SBEII-1 or Wheat SBEII-2 fragments) were radiolabelled using the Rediprime 11™ system (Amersham International) using α^{32}PdCTP (Amersham International) according to manufacturers instructions. Blots were hybridized overnight at 65°C in 0.6M NaCl, 20mM Pipes, 4mM $Na_2EDTA2H_2O$, 0.2% gelatin, 0.2% Ficoll 400, 0.2% PVP-360, 10mM $Na_4P_2O_710H_2O$, 0.8% SDS, 0.5mg/ml denatured salmon sperm DNA. Post hybridization washes were carried out in 30mM NaCl, 2Mm $NaH_2PO_4.2H_2O$, 0.2mM $Na_2EDTA.2H_2O$, 0.1% SDS at room temperature for 7 minutes, then 65°C for 10 minutes. Filters were exposed to Kodak BioMax MR™ (Amersham International) film at -70°C. Blots were stripped by washing in 15mM NaCl, 1mM $NaH_2PO_4.2H_2O$, 0.1mM EDTA at 90°C for 10 minutes, or until no counts above background remained.

Extension of the SBEII-1 3' sequence towards the 5'end of the mature peptide

We have exploited the sequence divergence between our wheat SBEII-1 and SBEII-2 sequences to design the SBEII-1 specific 3' primer, Sb4. This primer was used in conjunction with an SBEII specific 5' primer to extend the novel SBEII-1 sequence using a PCR-based approach.

To extend the SBEII-1 3' sequence towards the 5'end of the mature peptide, a second conserved domain was identified and an oligonucleotide sense primer, AGSBEI, designed. PCR amplification from the endosperm first strand cDNA pool was carried out using the AGSBEI-Sb4 primer pair. Separation of the amplification products by electrophoresis through a 1% (w/v) agarose gel (data not shown) showed that the reaction yielded a distinct band of approx. 2.2kb. The approx 2.2kb amplification products were excised from the gel, ligated with PT7Blue and transformed into competent Novablue *E.coli* cells. Following overnight culture, nine putative recombinant clones were selected for further analysis. Screening of each of the selected clones using vector specific primers indicated that clones 5A1, 5A2, 5A5 and 5A9 harboured inserts of the predicted size. Of these clone 5A1 (which falls in sub-class C) was selected for sequencing (Figure 10). The amino acid sequence of Figure 10 corresponds to the OsbeII-1ALL sequence of Figure 2. Although not full length the predicted open reading frame includes nucleotides 44 through to 1823 and encodes a 593 amino acid peptide. Based on similarities with the maize genes, it is estimated that this sequence is missing approximately 230 amino acids out of a predicted total of approximately 830 amino acids. On this basis, the partial sequence represents about 70% of the coding sequence. Multiple sequence alignment of this SBEII-1 sequence with recently published wheat SBEII-2 (Nair *et al.*, 1997), SBEI (Rapellin *et al.*, 1997) and SBEI-D2 (Rahman *et al.*, 1997) sequences showed that the SBEII-1 sequence has similarity indices of 69.6%, 31.2% and 46.7% to SBEII-2, SBEI and SBEI-D2 respectively (Figures 11 and 11a). This demonstrates that the SBEII-1 sequence differs from the published wheat SBE sequences, and confirms the analysis of the 3' sequence alignment (Figure 3). The increase in relative homology when compared to the values obtained following 3'sequence alignment results from the fact that the central domain of SBEs is highly conserved (Burton *et al.*, 1995; Gao *et al.*, 1997). However, it is clear

that this cloned wheat SBEII-1 sequence is significantly different from previously published wheat SBE sequences and represents a novel sequence.

Full experimental details were as follows.

SBEII-1 sequences were extended toward the 5'end of the mature peptide by amplification from the endosperm cDNA pool using the SBEII-1 specific primer Sb4 [TTTTCTTCACAACGCCCTGGG] (SEQ ID No: 40) in conjunction with the primer AGSBEI [TGTTTGGGAGATCTTCCTCCC] (SEQ ID No: 41). AGSBEI was designed to be homologous to the GVWEIFLP (SEQ ID No: 42) motif which is conserved in all known SBE sequences and is situated toward the 5'end of the mature peptide coding sequence. PCR was carried out in a 50μl reaction, comprising 5μl of the cDNA pool, 50pmol Sb4, 50pmol SBEA1, 5μl 5x Taq buffer, 4μl 25mM Mg^{2+}, 0.5μl 20mM dNTPs, and 1.25u Taq polymerase. All of the reaction components were mixed, before thermocycling at (94°C-45sec, 55°C-30sec, 72°C-1min 30sec) x 30 cycles, followed by a final 10 min extension step at 72°C. Amplification products were separated by electrophoresis through a 1%(w/v) agarose gel and specific amplification products of the predicted size were excised from the gel. The DNA was eluted from the gel slice using QIAGEN's gel extraction kit according to the manufacturers recommendations before ligation with pT7 Blue (Novagen). Ligation was carried out in a 10μl reaction volume comprising 7.5μl purified amplification product, 1μl 10x ligation buffer, 1μl pT7Blue and 0.5μl T4 DNA ligase (Amersham). The reaction components were mixed well before being placed at 4°C overnight. Following overnight incubation, half of the ligation reaction was used to transform competent Novablue *E.coli* cells (Novagen). Transformed cells were plated out onto LB plates supplemented with X-gal (40μgml^{-1}), IPTG (0.1mM), Carbenicillin (100μgml^{-1}), and Tetracycline (12.5μgml^{-1}), before placing at 37°C overnight. Putative recombinant clones were initially screened for the presence of an insert by colony PCR using the vector specific primers T7B and U19. Insert positive clones were then screened using an insert specific primer in conjunction with either T7B or U19 primers to determine the orientation of the insert within the multiple cloning site prior to sequencing.

Southern blot analysis

Southern analyses of the pre-made nulli-tetra and ditelosomic blots were carried out essentially as described in Jack *et al* (1994).

The SBEII-1 clones discussed above have been cloned into transformation vectors for transformation of wheat.

Northern blot analysis

Northern blots were prepared from total RNA from developing wheat grains of the cultivar Bobwhite. Figure 12 shows a northern blot of RNA from wheat grains of the cultivar Bobwhite grown in the glasshouse as described and harvested between 5 and 29 days after anthesis. The blot was probed with the 1kb Sac1 SBEII-1 fragment and subsequently (following blot stripping) with the 1.2kb BamH1 SBEII-2 fragment, both fragments purified and labelled as described. In Figure 12 panel A shows the Ethidium Bromide-stained RNA gel prior to northern transfer. Panel B shows the results of probing with the SBEII-1 probe and panel C shows the results of probing with the SBEII-2 probe. Comparing within and between panels B and C differences can be observed in the relative intensities of the signals at the different time points. In particular a relatively stronger signal intensity is observed with the SBEII-2 probe for the 5 day time point than with the SBEII-1 probe, indicating that the transcript profiles for SBEII-1 and SBEII-2 are distinct, suggesting that the two gene families (SBEII-1 and SBEII-2) are differentially expressed during grain development. The size of the transcripts observed for both SBEII-1 and SBEII-2 is approximately 3.5kb. However the SBEII-2 transcript is slightly smaller than the SBEII-1 transcript.

Plasmid constructions

Standard molecular biology procedures (Sambrook *et al*, 1989) were used for plasmid constructions.

pWxGS+ (Figure 13) comprising a maize granule bound starch synthase gene (Shure *et al* 1983) promoter-GUS-Nos fusion was obtained as a gift to Unilever Research from Sue Wessler (University of Georgia, Athens, USA) and may be obtained on request from that source. The promoter in pWxGS+ is approximately 1.5kb in length and represents a truncated version of a similar, but larger promoter fragment described in Russell & Fromm (1997). The sequence of the promoter (HindIII - BamH1 fragment) in pWxGS+ is presented in Figure 13A (SEQ ID No: 55).

pSRWXGUS1 (Figure 14) was produced by inserting a Sac 1 linker [d(pCGAGCTCG)0] (New England Biolabs [NEB]) (NEB catalogue No 1044) into the SmaI site in pWxGS+.

pVTWXGUS2 (Figure 15) was produced by inserting a BamH1 linker [d(pCGGGATCCCG)] (SEQ ID No: 43) (NEB catalogue No. 1071) into the Ecl136II (an isoschizomer of Sac1 which gives blunt ends) site of pWxGS+

A Sac1 linker was inserted at the XbaI site (which had been blunted using Klenow + dNTps) of the SBEII-1 Clone B6 in the plasmid pT7Blue to produce an intermediate clone. The SBE sequence was then purified from this intermediate clone as a Sac1 fragment and ligated into the Sac1 sites of pSRWXGUS1 replacing the GUS gene sequence to produce the plasmids pSR96-26 and pSR96-29 representing antisense and sense orientations of the SBEII-1 sequence downstream of the Waxy promoter, respectively.

A BamH1 linker was inserted at the XbaI site (which had been blunted using Klenow + dNTps) of the SBEII-2 Clone B11 in pT7Blue to produce an intermediate clone. The SBE sequence was then purified from this intermediate as a BamH1 fragment and inserted into the BamH1 sites of pVTWXGUS2, replacing the GUS gene sequence, to produce the plasmids pVT96-50 and pVT96-53 representing antisense and sense orientations, respectively, of the SBEII-2 sequence downstream of the Waxy promoter.

pVT96-54. A BamH1 linker was inserted at the Xba1 site (which had been blunted using Klenow + dNTPs) of the SBEII-2 clone B9 (equivalent to clone B1) in pT7Blue to produce an intermediate clone. The SBEII-2 sequence was then purified from this

intermediate clone as a BamH1 fragment and inserted into the BamH1 sites of pVTWXGUS2, replacing the GUS gene sequence, to produce the plasmid pVT96-54.

The Waxy-SBE-NOS sequences in the plasmids pSR96-26 and pSR96-29 and pVT96-50 and pVT96-53 were purified as HindIII/EcoRI fragments and inserted into the EcoRI/HindIII sites of plasmid pPBI-97-2 (also known as p97-2) (Figure 16). Plasmid pPBI-97-2 is described in European Patent Application No. 97305694.8 (published as WO 99/06570). Following removal of the ampicillin resistance marker gene the resulting plasmids were designated pSR97-26A- (clone B6 (SBEII-1, sub-class A) in antisense orientation), pSR97-29A- (clone B6 in sense orientation), and pSR97-50A- (clone B11 (SBEII-2, sub-class A) in antisense orientation) and pSR97-53A- (clone B11 in sense orientation) as illustrated in Figures 17, 18, 19 and 20, respectively.

p97-2C (Figure 21) was produced by digesting the polylinker sites Ecl136 II to SmaI in the plasmid pPBI97-2 (Figure 16), ligating and selecting recombinants in which the polylinker region from SmaI to Ecl136 II had reinserted in the opposite orientation.

The Waxy-NOS sequences in pSRWXGUS1 were transferred as a HindIII/EcoRI fragment into the HindIII/EcoRI sites of plasmid p97-2C to produce the plasmid p97-2CWT1 (Figure 22).

pSC98-1 and pSC98-2. The 5' extended SBEII-1 clone 5A1 in pT7Blue (comprising SBE sequence from coordinate 43 to 2003bp in Figure 10) was digested with EcoRI and XbaI, followed by 'in-fill' of overhangs using Klenow polymerase and dNTPs. The resulting blunt ended SBE fragment was gel purified and ligated to p97-2CWT1 (Figure 22) which had been digested with Ecl136II and dephophorylated using calf intestinal phosphatase. The resulting recombinants were screened by restriction digest anaylsis and clones comprising both orientations of the SBE sequence (with respect to the waxy promoter) were identified. pSC98-1 (Figure 23) is an antisense version and pSC98-2 (Figure 24) is a sense version. Following removal of the ampicillin marker gene the resulting plasmids were designated pSC98-1A- and pSC98-2A- respectively.

Ubiquitin promoter - NptII selection construct:pUN1

pUN1 was made in the following way:

A SacI linker was inserted at the SmaI site of the plasmid pAHC25 (Christensen and Quail 1996) to produce an intermediate plasmid. The GUS gene was removed from this intermediate plasmid by digesting with SacI followed by self ligation and identification of recombinant molecules lacking the GUS sequence to produce the plasmid pPBI95-9. pPBI95-9 was digested with EcoRI and following self ligation recombinant molecules lacking the Ubi-BAR sequences were identified. The resulting plasmid is designated pPBI96-23. An NptII sequence was amplified as a PCR product using the primers AG95-7:

5'GATGAGCTCCGTTTCGCATGATTGAACAAGATGG (SEQ ID No: 44) and AG95-8: 5'GTCGAGCTCAGAAGAACTCGTCAAGAAGGC (SEQ ID No: 45), using pPBIBAG3 (Goldsbrough *et al* 1994 as template for the NptII sequence. The amplified product was cloned into the SstI site of pBluescript (Stratagene) and sequenced. The sequencing revealed that the NptII sequence was of the 'mutant' form rather than the wild-type as had been expected. The 'mutant' form carries a single base change which is flanked by unique Nco1 and Sph1 sites. The pBluescript clone was digested with Nco1 and Sph1 to remove the region containing the single base change. Two oligonucleotides, (Npt1:CCCGACGGCGAGGATCTCGTCGTGACC (SEQ ID No: 46) and Npt2: CATGGGTCACGACGAGATCCTCGCCGTCGGGCATG) (SEQ ID No: 47) were then annealed to each other to form an Nco1/Sph1 fragment. This was cloned into the Nco1/Sph1 digested Bluescript/Npt11 clone, and the resulting clone was sequenced to confirm that the gene was now of the wild type form.

The NptII sequences was then purified as a Sac1 fragment and inserted at the SacI site of pPBI96-23 to produce pUN1 (Figure 25). pUN1 includes the wild-type ubiquitin promoter (Ubi promoter), which is also referred to as the ubiquitin regulatory system (abbreviated to URS). The orientation of the NptII sequence in pUN1 was determined by restriction digest analysis. The sequence of the NptII Sac1 fragment is presented in Figure 26 (SEQ ID No: 35).

pUSN99-1 and pUSN99-2. The SBEII-1 (clone B6) sequence was purified as a Sac1 fragment from the plasmid pSR96-26 and inserted at the Sac1 site of pPBI96-23 to produce the plasmids pUSN99-1 and pUSN99-2 (Figures 27 and 28) representing sense and antisense orientations of the SBEII-1 sequences respectively.

pPBI97-2BdUN1. pPBI92-2BdUN1 (also sometimes referred to as p97-2BdUN1) comprises a reconstituted ubiquitin regulatory system (referred to hereafter as a modified ubiquitin promoter or a modified ubiquitin regulatory system (mURS)) which lacks the two overlapping 'consensus heatshock elements' discussed in EP 0342926 and US 5614399. The modified ubiquitin promoter was prepared via PCR amplification of two DNA fragments using maize genomic DNA as template, followed by ligation of the two fragments to produce a single fragment lacking the consensus heatshock (HS) elements. A Kpn1 restriction site was engineered in place of the HS elements. The primers used were designed from sequence information published by Liu et al 1995 (EMBL DNA database accession ZMU29159). To delete the HS elements and to replace with a diagnostic Kpn1 site the ubiquitin promoter and intron sequences were amplified as two fragments using the primer combinations HS1 + Ubi3-3 and HS2 + Ubi5-2, the sequences of which are given below. Primers Ubi5-2 and Ubi3-3 are homologous to sequences in the sequence published by Liu et al 1995. Primers HS1 and HS2 are homologous to sequences located immediately 3' and 5' respectively of the two overlapping HS elements in the ubiquitin promoter as described in EP 0342926 and US 5361399. Both of these primers have a Kpn1 tail at their 5' ends.

<u>Primers</u>

HS1: 5-ATTAGGTACCGGACTTGCTCCGCTGTCGGC - 3 (SEQ ID No: 48)
HS2: 5-TATAGGTACCGAGGCAGCGACAGAGATGCC -3 (SEQ ID No: 49)
Ubi5-2: 5-AGCTGAATCCGGCGGCATGGC -3 (SEQ ID No: 50)
Ubi3-3: 5-TGATAGTCTTGCCAGTCAGGG -3 (SEQ ID No: 51)

The amplified products were subcloned into pGEM TEasy (Promega) to produce the plasmids p97-U1 and p97-U2. The full-length (approx. 2Kb) modified ubiquitin promoter

was reconstructed by subcloning the Kpn1 - Sac1 fragment from p97-U1 into the Kpn1/Sac1 sites of p97-U2 to produce p97-U3. A partial restriction map of the predicted sequence (SEQ ID No: 52) of the cloned fragment in p97-U3 is presented in Figure 29. (The modified ubiquitin promotor (or mURS) is the subject of a copending European Patent Application filed by the present applicants on the same day as the present application, under the reference C1235.01/M). The modified ubiquitin promoter was transferred as a PstI fragment from p97-U3 into plasmid pPBI96-36. The plasmid pBI96-36 (Figure 30) comprises the GUS-Nos reporter gene fusion under the control of the wild-type ubiquitin promoter (derived from pAHC25) in a pUC plasmid backbone. The promoter replaces the wild-type ubiquitin regulatory system in pPBI96-36 to produce an intermediary plasmid p97-dUG1 (Figure 31).

Construction of pPBI97-2BdUN1

The Ubi-Nos sequences in pPBI96-23 were transferred as an EcoRI - HindIII fragment into the EcoRI and HindIII sites of p97-2B (plasmid p97-2B is described in European Patent Application No. 97305694.8 published as WO 99/06570) to produce the plasmid p97-2BUbiNos. The modified ubiquitin promoter was purified as a HindIII/SacI fragment from p97-dUG1 (Figure 31) and transferred into the HindIII and SacI sites of p97-2BUbiNos, replacing the wild-type ubiquitin promoter to produce p97-2BdUbiNos. The NptII sequence in pUN1 was purified as a SacI fragment and transferred into the SacI site of p97-2BdUbiNos to produce pPBI97-2BdUN1 (Figure 32). Following removal of the ampicillin resistance marker using the method as described in WO 99/06570, the resulting plasmid as used for wheat transformation was designated p97-2BdUN1A-

pCaineo

pCaiNeo comprises the NptII gene under control of a CaMV35S promoter and maize Adh1 intron. The plasmid is described in Fromm et al 1986.

Transformation of wheat

The following plasmid combinations (co-bombardments) have been used in the transformation of wheat plants:

Table 2. Plasmid combinations used in wheat transformation experiments.

Starch gene construct/s	Selection marker construct
	pAHC25
pWXGS+	pUN1
pSR97-26A- antisense	pUN1 or p97-2BdUN1
pSR97-29A- sense	p97-2BdUN1 or pCaiNeo
pSC98-1A- antisense	p97-2BdUN1
pUSN-1 sense	p97-2BdUN1
pUSN-2 antisense	p97-2BdUN1
pUSN-1 sense & pUSN-2 antisense	pUN1
pSC98-2A- sense	p97-2BdUN1

The wheat transformation methods used and described here are largely based on those described by Barcelo and Lazzeri, 1995.

Embryo wheat plants of the spring cultivar Bobwhite and the winter cultivar Florida were grown in a glasshouse with 16hr day length supplemented with lights to maintain a minimum light intensity of 500 umol m-^{2}s-^{1} at 0.5M above flag leaf. Glasshouse temperatures were maintained at 19°C+/-1°C during the day and 14°C+/-1°C at night.

Immature embryos of wheat were harvested from developing grain. The seeds were harvested and embryos were cultured at approximately 12 days after anthesis when the embryos were approximately 1mm in length. Seeds were first rinsed in 70% ethanol for 5 minutes and then sterilised in a 10% solution of Domestos bleach (Domestos is a Trade

Mark) for 15 minutes followed by 6 washes with sterile distilled water. Following removal of the embryonic axis the embryos were placed axis surface face down on agargel (Sigma catalogue no. A-3301) solidified MM1 media. The general recipe for MM1 is given in Appendix 1, and the recipes for the various constituents in Appendix 2. The embryos were maintained in darkness for one to two days at 24°C +/-1°C prior to bombardment.

The plasmids pAHC25, pCAiNeo, pUN1 and p97-2BdUN1 were used to provide selection markers in the combinations with starch gene constructs as detailed in Table 2. pAHC25 (Christensen and Quail 1996) contains a chimeric Ubi-BAR gene which provides selection of transformants to phosphinothricin, the active ingredient in herbicides BASTA™ and Bialophos (see Block, M.de. *et al* 1987). The plasmids pCAiNeo (Fromm *et al.*, 1986), pUN1 and p97-2BdUN1 contain chimeric promoter-NptII gene fusions and provide selection of transformants against a range of aminoglycoside antibiotics including kanamycin, neomycin, geneticin and paromycin.

Particle bombardments was used to introduce plasmids into plant cells. The following method was used to precipitate plasmid DNA onto 0.6μm gold particles (BIO-RAD catalogue number 165-2262): A total of 5μg of plasmid DNA was added to a 50μl sonicated for one minute suspension of gold particle (@ 10mg/ml) in a 1.5ml microfuge tube. Following a brief vortex for three seconds 50μl of a 0.5M solution of calcium chloride and 20μl of a 0.05M solution of spermidine free base were added to the opposite sides of the microfuge tube lid. The tube contents were mixed together by closing the lid and tapping the calcium chloride and spermidine to the bottom of the tube. Following a vortex for three seconds the suspension was centrifuged at 13,000 rpm for 5 seconds. The supernatant was then removed and the pellet resuspended in 150μl of absolute ethanol. This requires scraping the gold particles off the inside of the tube using a pipette tip. Following a further three second vortex, the sample was centrifuged again and the pellet resuspended in a total volume of 85μl in absolute ethanol. The particles were vortexed briefly and sonicated for 5 seconds in a Camlab Trisonic T310 water bath sonicator to ensure fine dispersion. An aliquot of 5μl of the DNA coated gold particles were placed in the centre of a macrocarrier (BIO-RAD catalogue no. 115-2335) and allowed to dry for

30 mins. Particle bombardment was performed by using a Biolisitc™ PDS-1000/He (BIO-RAD Instruments, Hercules CA) chamber which is illustrated schematically in Figure 33, using helium pressure of 650 and 900 psi (rupture discs: BIO-RAD catalogue numbers 165-2327 and 165-2328 respectively).

Referring to Figure 33, the illustrated vacuum chamber comprises a housing 10, the inner side walls of which include a series of recesses 12 for receiving shelves such as sample shelf 14 shown at the fourth level down from the top of the housing. A rupture disc 16 is supported in a He pressure shock tube 18 near the top of the housing. A support 20, resting in the second set of recesses 12 down from the top of the housing, carries unit 22 that includes a stopping screen and a number of rings 24, with 11 rings below the support 20 and 3-4 rings above the support 20. Macrocarrier 26 is supported at the top of unit 22. The approximate distance from the rupture disc 16 to the macrocarrier 26 is 25mm, with the approximate distance from the macrocarrier 26 to the stopping screen being 7mm, and the approximate distance from the stopping screen to the sample shelf 14 being 67mm. The top of unit 22 is about 21mm from the bottom of the shock tube 18, and the bottom unit 22 is about 31mm from the top of sample shelf 14.

Immature embryos were bombarded between 1 and 2 days after culture. For bombardment the immature embryos were grouped into a circular area of approximately 1cm in diameter comprising 20-100 embryos, axis side face down on the MM1 media. The Petri dish (not shown) containing the tissue was placed in the chamber on shelf 14, on the fourth shelf level down from the top, as illustrated in Figure 33. The air in the chamber was then evacuated to a vacuum of 28.5 inches of Hg. The macrocarrier 26 was accelerated with a helium shock wave using rupture membranes that burst when the He pressure in the shock tube 18 reaches 650 or 900 psi. Within 1 hour after bombardment the bombarded embryos were plated on MM1 media at 10 embryos per 9cm petri dish and then maintained in constant darkness at 24°C for 2-3 weeks. During this period somatic embryogenic callus was produced on the bombarded embryos.

After 2-3 weeks the embryos were transferred onto agar-solidified regeneration media, known as R media, and incubated under 16hr daylength at 24°C. The general recipe for

R media is given in Appendix 1. Embryos were transferred on fresh plates at 2-3 week intervals. The composition of the regeneration media varied depending on which selection regime was to be used. For transformants bombarded with the BAR gene the 3 amino solution was omitted and PPT (phosphinothricin) at 1mg/L, rising to 3mg/L over a period of three 2-3 week transfers was used for selection. For selection of transformants using the NptII gene three different regimes were used: 1) Geneticin (GIBCO-BRL catalogue no. 10131-019) was incorporated (at 50mg/L) immediately on transfer to regeneration media and maintained at 50mg/L on subsequent transfers to regeneration media. 2) & 3) Embryos were first transferred to regeneration media without selection for 12 days and 2-3 weeks, respectively, and thereafter transferred on to media containing Geneticin at 50mg/L. After 2-3 passages on regeneration media regenerating shoots were transferred to individual culture tubes containing 15 ml of regeneration media at half salt strength with selection at 3mg/L PPT or 35mg/L geneticin depending on whether the BAR gene of NptII gene had been used in the original bombardments. Following root formation the regenerated plants were transferred to soil and the glasshouse.

Genomic DNA isolation and Southern Analyses

Southern analyses of primary transformants and progeny material were carried out as follows: Freeze dried leaf tissues were ground briefly in a Kontes™ pestle and mortar, and genomic DNA extracted as described in Fulton et al, 1995. 5 μg of DNA were digested with an appropriate restriction enzyme according to the manufacturers instructions, and electrophoresed overnight on a 1% agarose gel, after which the gel was then photographed, washed and blotted onto Hybond N+ ™ (Amersham International) according to the method of Southern using standard procedures (Sambrook et al 1989). Following blotting, the filters were air dried, baked at 65°C for 1-2 hours and UV fixed at 312nm for 2 minutes.

Probe preparation and labelling for the Southern analyses of transformed material was carried out as described above.

GUS histochemistry was performed essentially as described in Jefferson (1987).

Evaluation of the ubiquitin promoter for constitutive expression of associated transgenes.

The plasmid pAHC25 (Christensen and Quail, 1996) was transformed into wheat as described in previous sections. Transformants were selected on the basis of resistance to phosphinothricin. Southern blot analyses were carried out on the primary transformants to confirm integration of the plasmid sequences (data not shown). GUS histochemical analyses were also carried out and demonstrated that the ubiquitin promoter is capable of mediating high levels of GUS expression in a range of wheat tissues. Figure 34 A, B, C & D show histochemical localisation of GUS expression in the seed, stem, floral and leaf tissues respectively. Southern blot and GUS histochemical analyses were also carried out on self progeny from primary transformants to confirm that the transformation system used is capable of producing transgenic plants which stably transmit the integrated plasmid sequences to progeny plants. Figure 35 shows a Southern blot of 26 progeny plants of transformant BW119 which had been transformed with pAHC25. In this example genomic DNA from the progeny plants was digested with the restriction enzyme Sac1 and the blot was probed with the GUS gene coding sequence. The Southern blot results are suggestive of the presence of two independently segregating integration loci, each comprising concatamers of pAHC25 plasmid sequences.

Evaluation of the maize waxy promoter for endosperm-specific expression of associated transgenes.

The plasmids pWxGS+ and pUN1 were co-transformed into wheat as described in previous sections. Transformants were selected on the basis of resistance to geneticin. Southern blot analyses were carried out on the primary transformants to confirm integration of the plasmid sequences (data not shown). Gus histochemical analyses were also carried out to determine the expression profile mediated by the maize waxy promoter. The majority of the transformants that expressed GUS exhibited expression specifically in endosperm tissue, demonstrating the suitability of this promoter for mediating endosperm expression of associated transgenes. Figure 36 A & B shows endosperm specific expression of GUS in seeds from two independent transformants. We did not observe GUS expression in pollen grains as was seen by Russell and Fromm (1997), however the

construct they used also incorporated the maize hsp 70 intron which may conceivably have influenced expression both quantitatively and qualitatively.

Transformation of wheat with starch gene constructs.

The various construct combinations detailed in Table 2 were co-transformed into wheat using the procedures as described in previous sections. Transformants were selected on the basis of resistance to geneticin. The primary transformants were confirmed positive by Southern blot analysis. Blots were sequentially probed with an NptII coding sequence probe and a SBE coding region probe. Figure 37 shows an example of a Southern blot which comprises 22 putative transformants which had been co-bombarded with pSR97-29A- or pSR97-26A- and pUN1 or p97-2BdUN1. Genomic DNAs on this blot had been digested with Sac1. The blot was first probed with the NptII probe. Lanes marked with an asterisk correspond to transformants which give a positive signal with the NptII probe. The blot shown in Figure 37 was probed with the SBEII-1 1kb Sac1 fragment. The Sac1 digest is expected to release a 1kb SBEII-1 hybridising band from both pSR97-29A- and pSR97-26A- plasmid sequences, and the intensity of this band will vary depending on the copy number of inserted plasmid sequences. As can be seen in Figure 37 several additional SBEII-1 hybridising bands are also observed. Five of these bands are present in all lanes and result from hybridisation to endogenous wheat SBEII-1 sequences. The additional bands of varying size which are observed in the majority of lanes which show the 1kb hybridising band most likely result from integration events in which one or more copies of the plasmid had been linearised within the 1kb SBEII-1 sequence prior to integration. In the example shown in Figure 37, of the 20 NptII positive plants, 16 were found to be co-transformed with the SBEII-1 sequences, representing a co-transformation efficiency of 80%.

Differential Scanning Calorimetry (DSC)

When heated, an aqueous suspension of starch in excess water undergoes a co-operative endothermic transition known as gelatinisation, as discussed above, entailing a melting of the starch crystallites. Differential scanning calorimetry (DSC) measures the amount of

energy (heat) absorbed or released by a sample as it is heated, cooled or held in a constant (isothermal) temperature. DSC has been widely used to study the gelatinisation and retrogradation of starch.

DSC analyses were carried out on single grains or pools of 5 grains from primary transformants generated through transformation using each of the gene construct combinations detailed in Table 2.

Two different sample preparation and DSC methodologies were used:

Method 1:

Individual seed samples were crushed and ground using a pestle and mortar. The resulting bran was then separated and samples weighed into 50μm aluminium DSC pans. Water, three times by weight, was added and the sample pans sealed. Analyses were performed using a Perkin-Elmer DSC-7 Robotic™ system equipped with an Intercooler II™, for sub-ambient conditions. Samples were heated from 25°C to 80°C at a heating rate of 5°C min^{-1}. Gelatinisation enthalpy, onset and peak and end temperatures were recorded. The thermograms were analysed using the Perkin-Elmer software programs (Thermal Analysis Software 7). Gelatinisation enthalpy is expressed in Joules (J)/gram (g) of sample.

Method 2:

Pools of 5 seeds from a single primary transformant, or single seeds from primary transformants, were milled using a Cemotec 1090™ Sample Mill. The milled sample was then passed through a 250 micron sieve to separate the bran from endosperm. Approximately 5mg of the sieved samples was then accurately weighed into 50μl aluminium DSC pans. Water, three times by weight, was added and the sample pans sealed. Analyses were performed using a Perkin-Elmer Pyris 1™ DSC equipped with autosampler and Intracooler IP. Samples were heated from 40°C to 85°C at a heating rate of 10°C per minute. The thermograms were analysed using the Perkin-Elmer software programs (Pyris Software for Windows v 3.5). Gelatinisation enthalpy, onset and peak

and end temperatures were recorded.

Using method 1, DSC analyses were performed on individual mature grains of primary transformants, transformed with the plasmid combinations pSR97-26A-/pUN1, pSR97-26A-/p97-2BdUN1 and pSR97-29A-/p97-2BdUN1. Data obtained were compared to data from control material which had been transformed with one of the NptII selectable marker plasmids, but did not contain any of the 'starch' plasmids. Table 3 summarises the average onset, peak, end and enthalpy values for the selected material. The majority of samples showed similar values to the control material. However, as can be seen from Table 3 onset, peak and end temperatures were higher for a number of the transgenic samples compared to the control material. For example, transformant BW 326 exhibits a 6.7°C, 4.9°C and 4.6°C increase in onset, peak and end temperatures (respectively) compared to the control sample.

Using method 2 a further series of DSC analyses were carried out on pools of 5 grains from primary transformants, transformed with the plasmid combinations pSC98-1A-/p97-2BdUN1, pUSN-1/p97-2BdUN1, pUSN-2/p97-2BdUN1 and pUSN-1/pUSN-2/pUNI. Data obtained were compared to data from control material which had been transformed with one of the NptII selectable marker plasmids, but did not contain any of the 'starch' plasmids. Table 4 summaries the onset, peak, end and enthalpy values for the selected pooled samples. In many cases there is evidence that the 'starch' transgenic material shows onset, peak and end temperatures which are greater than those observed for the control material. For example, transformant BW727 exhibits a 9.8°C, 8.7°C and 9.1°C increase in onset, peak and end temperatures (respectively) compared to the BW control sample 3, and a 7.6°C, 6.8°C and 7.8°C increase in onset, peak and end temperatures (respectively) compared to the BW control sample 2.

Table 3: Results of DSC analyses on single grains using method 1. Data shown are the averages of between 2 and 6 individual grain samples (T_o, T_p and T_f are onset, peak and end temperatures respectively).

Plasmid combination	Line Code	T_o (°C)	T_p (°C)	T_f (°C)	ΔH (J/g)
BW control sample 1		55.2	59.7	66.5	4.66
pSR97-26A-/pUN1	BW283	57.1	60.4	65.0	2.12
	BW135	57.2	62.1	68.6	4.86
	BW324	57.8	62.1	69.1	5.33
	BW325	58.4	61.8	68.7	3.90
	BW326	61.9	64.6	71.1	2.46
	BW348	60.7	63.4	69.7	3.76
pSR97-26A-/p97-2BdUN1	F227	57.4	61.4	67.3	2.65
pSR97-29A-/p97-2BdUN1	F310	62.1	63.7	69.2	6.75
	F312	59.0	62.3	66.8	1.16
	BW335	56.2	60.8	69.1	4.63
	BW353	59.5	62.7	70.8	3.21
	BW354	55.4	61.7	68.9	4.28
	BW355	57.9	61.5	68.0	3.95
	BW357	55.3	60.6	68.0	3.74
	BW363	56.7	62.5	67.9	1.13
	BW367	59.0	62.5	68.2	2.17
	BW369	57.9	60.9	65.9	1.04
	BW370	53.7	59.4	67.5	6.00
	BW375	57.2	61.5	70.0	4.14
	BW376	54.0	58.1	68.0	3.39
	BW377	53.4	60.9	69.2	2.60
	BW380	54.6	61.6	67.6	2.16
	BW390	56.8	61.2	68.5	1.29
	BW399	57.4	62.7	67.9	1.77
	BW400	60.6	63.6	68.1	0.64
	BW341	51.6	59.0	66.4	1.97

Table 4: Results of DSC analyses on pools of 5 grains using method 2. T_o, T_p and T_f are onset, peak and end temperatures respectively

Plasmid combination	Line Code	T_o (°C)	T_p (°C)	T_f (°C)	ΔH (J/g)
F control sample 1		60.1	63.9	68.0	6.30
BW control sample 2		59.3	64.0	68.4	5.94
BW control sample 3		57.08	62.09	67.08	4.28
pSC98-1A-/p97-2BdUN1	BW449	59.3	62.9	67.9	3.95
	BW477	57.7	63.6	70.6	8.30
	F492	62.3	66.4	70.2	7.60
	F494	63.6	67.3	71.0	5.73
	BW511	59.6	63.8	67.2	0.98
	BW518	60.2	64.9	69.2	3.57
	BW519	58.4	63.6	68.5	4.13
	BW527	58.7	63.7	69.0	6.38
	BW549	59.9	64.8	69.3	4.48
	BW550	60.2	64.6	68.9	5.06
	BW552	60.8	62.9	67.9	3.74
	BW553	59.5	63.9	67.5	3.60
	BW555	61.0	66.1	68.2	5.43
	BW557	62.7	66.9	71.0	5.08
	BW559	61.6	65.9	70.8	5.08
	BW563	61.4	65.1	69.4	1.90
	BW564	59.4	64.5	73.2	7.08
	BW576	61.8	65.6	69.3	2.65
	BW587	61.3	65.4	69.4	5.36
	BW614	63.9	67.9	71.8	5.83

	BW618	61.3	65.6	69.7	3.54
	BW583a	58.9	63.7	68.0	3.54
	BW631	61.5	65.6	69.7	4.52
	BW633	61.9	66.0	70.2	5.12
	BW634a	60.8	64.9	70.2	5.10
	BW637a	62.8	67.2	72.0	5.16
	BW639	61.8	65.1	68.9	2.15
	BW640a	62.2	66.7	71.0	3.23
	BW642	63.2	67.2	70.9	4.90
	BW698	62.9	67.0	70.9	4.48
	BW700a	63.8	67.6	71.2	3.41
	BE524a	59.4	64.3	68.9	4.05
pUSN-1/p97-2BdUN1	BW622	59.0	64.1	68.7	4.32
	BW628	56.2	63.3	66.0	6.09
	BW645	57.5	65.6	69.5	5.97
	BW646	61.6	66.4	67.7	3.99
	BW647	61.3	65.4	69.0	3.47
	BW648	59.8	64.4	68.8	4.65
	BW649	61.3	65.6	70.1	5.07
	BW656	59.9	64.6	69.2	5.38
	BW660	62.0	67.3	71.0	4.23
	BW661	61.5	65.8	69.6	3.88
	BW664	61.1	66.1	70.8	4.81
	BW665	61.6	66.5	69.4	5.25
	BW667	63.0	67.1	70.8	3.91
	BW672	63.0	68.1	71.9	5.43
	BW673A	63.1	67.7	71.6	4.83
	BW675	62.1	66.4	71.3	10.97

	BW676	59.8	67.3	71.2	4.21
	BW678	63.0	66.3	69.3	1.20
	BW680	60.8	65.3	70.1	4.94
	BW701	62.3	67.5	72.2	4.70
	BW706	63.0	67.3	71.3	4.94
	BW707	60.9	65.8	70.0	4.77
	BW708	61.7	65.5	68.8	6.11
	BW726	62.6	67.5	71.3	5.44
	BW755	60.8	65.8	70.6	5.18
	BW702	61.9	67.0	71.0	4.44
	BW756	62.3	66.1	69.7	4.83
pUSN-2/p97-2BdUN1	BW625	62.7	68.2	73.8	4.27
	BW653	60.4	65.3	70.1	6.52
	BW704	60.9	66.2	70.2	4.19
	BW718	61.3	66.9	71.2	4.15
	BW719	62.2	67.2	71.7	5.32
	BW722	64.8	67.5	70.0	2.14
	BW740	63.4	67.9	72.3	5.67
	BW741	62.6	66.9	70.5	5.30
	BW742	64.6	67.9	72.0	6.66
	BW752	62.3	66.3	70.0	4.63
pUSN-1/pUSN-2/pUN1	BW685	62.6	65.5	69.0	2.60
	BW686A	61.9	66.3	70.2	4.45
	BW714	63.0	67.6	71.3	3.53
	BW727	66.9	70.8	76.2	5.19
	BW728	62.0	66.3	70.4	5.70
	BW731	63.3	67.9	73.0	4.90

	BW732	63.5	66.8	70.8	4.11
	BW748	62.1	67.4	71.9	5.38
	BW794	62.8	67.5	71.8	5.17

Appendix 1.

Recipe for 2x concentrated MM1 media

Constituent	Volume of stock per litre of 2x concentrated media
Macrosalts MS (10X stock)	200ml
Microsalts L (1000x stock)	2ml
FeNaEDTA MS (100x stock) [Sigma catalogue F-0518]	20ml
Modified Vits MS (x1000)	1ml
3 amino acid solution (25x stock)	40ml
myo inositol (Sigma catalogue number I-3011)	0.2g
sucrose	180g
$AgNO_3$ (20mg/ml stock) Added after filter sterilisation	1ml
Picloram (1m/ml stock) Added after filter sterilisation	4ml

Filter sterilise and add to an equal volume of moulten 2x agargel (10g/L).

Recipe for 2x concentrated R media

Constituent	Volume of stock per litre of 2x concentrated media
Macrosalts L7 (10X stock)	200ml
Microsalts L (1000x stock)	2ml
FeNaEDTA MS (100x stock)	20ml
Vits/Inositol L2 (200x stock)	10ml
3 amino acid solution (25x stock)	40ml
Maltose	60g
2,4-D (1mg/ml stock) added after filter sterilisation	200μl
Zeatin cis trans mixed isomers (Melford labs catalogue no. Z-0917) (5mg/ml stock) added after filter sterilisation	2ml

Filter sterilise and add to an equal volume of moulten 2x agar (16g/litre)

Appendix 2

Recipes for constituents of MM1 and R media

Microsalts L (1000x stock)

	per 100ml
$MnSO_4.7H_2O$	1.34g
H_3BO_3	0.5g
$ZnSO_4.7H_2O$	0.75g
KI	75mg
$Na_2MoO_4.2H_2O$	25mg
$CuSO_4.5H_2O$	2.5mg
$CoCl_2.6H_2O$	2.5mg

Filter sterilise through a 22μm membrane filter

Store at 4°C

Macrosalts MS (10X stock)

	per litre
NH_4NO_3	16.5g
KNO_3	19.0g
KH_2PO_4	1.7g
$MgSO_4.7H_2O$	3.7g
$CaCl_2.2H_2O$	4.4g

NB: Dissolve $CaCl_2$ before mixing with other components

NB: Make up KH_2PO_4 separately in sterile H_20, and add last.

Store solution at 4°C after autoclaving

Modified MS Vits (1000x stock)

	Per 100ml
Thiamine HCl	10mg
Pyridoxine HCl	50mg
Nicotinic acid	50mg

Store solution in 10ml aliquots at -20°C.

3 amino acid solution (25x stock)

	Per litre
L-Glutamine	18.75g
L-Proline	3.75g
L-Asparagine	2.5g

Store solution in 40ml aliquots at -20°C

Macrosalts L7 (10x stock)

	per litre
NH_4NO_3	2.5g
KNO_3	15.0g
KH_2PO_4	2.0g
$MgSO_4.7H_2O$	3.5g
$CaCl_2.2H_2O$	4.5g

NB: Dissolve $CaCl_2$ before mixing with other components

NB: Make up KH_2PO_4 separately in 50ml H_20 and add last

Store solution at 4°C after autoclaving

Vits/Inositol (200x stock)

200x Stock	Per 100ml
Inositol	4.0g
Thiamine HCl	0.2g
Pyridoxine HCl	0.02g
Nicotinic acid	0.02g
Ca-pantothenate	0.02g
Ascorbic acid	0.02g

Store solution in 40ml aliquots at -20°C

REFERENCES

Atwell, W.A., Hood, L.F., Lineback, D.R., Varriano-Marston, E., and Zobel, H.F. (1988). The terminology and methodology associated with basic starch penomena. Cereal Foods World 33, 306-311.

Baba, T., Kimura, K., Mizuno, K., Etoh, H., Ishida, Y., Shida, O., and Arai, Y. (1991). Sequence conservation of the catalytic regions of amylolytic enzymes in maize branching enzyme-I. Biochem. Biophys. Res. Comm. 181(1): 87-94.

Barcelo P., and Lazzeri, P. (1995). Transformation of cereals by microprojectile bombardment of immature inflorescence and scutellum tissues. Methods in Molecular Biology - Plant Gene Transfer and Expression Protocols (vol 49), 113-123. Jones H [ed] Humana Press Inc., Totowa, NJ.

Block, M.de *et al.* (1987). EMBO J 6:2513-2518

Boyer, C.D. and Priess, J. (1978). Multiple forms of (1-4)-α-D-Glucan, (1-4)-α-D-Glucan 6-glycosyl transferase from developing *Zea mays* L. kernals. Carbohydrate Res 61:321-334.

Burton, R.A., Bewley, J.D., Smith, A.M., Bhattacharyya, M.K., Tatge, H., Ring, S., Bull, V., Hamilton, W.D.O., and Martin, C. (1995). Starch branching enzymes belonging to distinct enzyme families are differentially expressed during pea embryo development. Plant J 7:1-13

Christensen, A.H., and Quail, P.H. (1996). Transgenic research, 5:213-218.

Donovan, J.W. (1979). Phase transitions of the starch-water system. Biopolym. 18, 263-275.

Fisher, D.K., Boyer, C.D., and Hannah, L.C. (1993). Starch branching enzyme II from maize endosperm. Plant Physiol. 102:1045-1046.

Fisher, D.K., Gao, M., Kim, K-N., Boyer, C.D., and Guiltinan, M.J. (1996). Two closely related cDNAs encoding starch branching enzyme from *Arabidopis thaliana*. Plant Mol. Biol. 30:97-108

Fromm ME, Taylor LP & Walbot V. (1986). Stable transformation of maize after gene transfer by electroporation. Nature 319: 791-793,

Fulton, T.M., Chunwongse, J., and Tanksley, S.D. (1995). Miniprep Protocol for Extraction of DNA from Tomato and other Herbaceous Plants, Plant Molecular Biology Reporter, 13 (3), pages 225-227.

Gao, M., Fisher, D.K., Kim, K-N., Shannon, J.C., and Guiltinan, M.J. (1997). Independent genetic control of maize starch-branching enzymes IIa and IIb: Isolation and characterisation of a *Sbe2a* cDNA. Plant Physiol. 114: 69-78.

Gaun, H.P., and Preiss, J. (1993). Differentiation of the properties of the branching isoenzymes from maize (*Zea mays*). Plant Physiol. 102: 1269-1273.

Goldsbrough AP, Belzile F, Yoder JI (1994). Complementation of the tomato anthocyanin without (*aw*) mutant using the dihydroflavonol 4-reductase gene. Plant Physiology 105, 491-496.

Islam, A.K.M.R. (1983). Ditelosomic additions of barley chromosomes to wheat. *In* Proc. 6th Int. Wheat Genet. Symp. 233-238. Kyoto, Japan.

Jack, P.L., Dimitrijevic, T.A.F., and Mayes S. (1994) Assessment of nuclear, mitochondrial and chloroplast RFLP markers in oil palm (*Elaeis guineensis Jacq.)* Theor. Appl. Genet. 90:643-649.

Jefferson RA (1987). Assaying chimaeric genes in plants: The GUS gene fusion system. Plant Molecular Biology Reporter 5 (4) 387-405.

Kennedy, J.F. and Cabalda, V.M.B. (1991). Bioactive carbohydrates in food. Chemistry in Britain, November, 1017-1019, 1026.

Khoshnoodi, J., Ek, B., Rask, L., and Larsson, H. (1993). Characterisation of the 97 and 103kDa forms of starch branching enzyme from potato tubers. FEBS 332 (1,2):132-138.

Kossmann, J., Visser, R.G.F., Muller-Rober, B., Willmitzer, L., and Sonnewald, U. (1991). Cloning and expression anaylsis of a potato cDNA that encodes branching enzyme: evidence for co-expression of starch biosynthetic genes. Mol. Gren. Genet 230: 39-44.

Liu et al (1995). Biochem Cell Biol 73: 19-30.

Martin, C., and Smith, A.M. (1995). Starch biosynthesis. Plant Cell 7:971-985.

Matzke & Matzke (1995). Plant Physiol. 107, 679-685.

Mizuno, K., Kawasaki, T., Shimada, H., Satoh, H., Kobayashi, E., Okumura, S., Arai, Y., and Baba, T. (1993). Alteration of the structural properties of starch components by the lack of an isoform of starch branching enzyme in rice seeds. J.Biol. Chem. 268(25): 19084-19091.

Morell, M.K., Rahman, S., Abrahams, S.L., and Appels, R. (1995). The biochemistry and molecular biology of starch synthesis in cereals. Auist.J. Plant Physiol. 22: 647-660.

Morell, M., Blennow, A., Kosar-Hashemi, B., and Samuel, M.S. (1997). Differential expression and properties of starch branching enzyme isoforms in developing wheat endosperm. Plant Physiol. 113:201-208.

Mousley, C.G. (1994). PhD Thesis, Wye College, University of London.

Nair, R.B., Baga, M., Scoles, G.J., Kartha, K.K., and Chibbar, R.N. (1997). Isolation, characterisation and expression analysis of a starch branching enzyme II cDNA from wheat. Plant Sci. 122:153-163.

Nakamura, Y., Takeichi, T., Kawaguchi, K., and Yamanouchi, H. (1992). Purification of two forms of starch branching enzyme (Q-enzyme) from developing rice endosperm. Physiol. Plant 84: 329-335.

Nakamura, Y., and Yamanouchi, H. (1992). Nucleotide sequence of a cDNA encoding starch-branching enzyme, or Q-enzyme I, from rice endosperm. Plant Physiol. 99:1265-1266.

Preiss, J. (1991). Biology and molecular biology of starch synthesis and regulation. *In* Oxford Surveys Plant Mol. Cell Biol. 7:59-114.

Rahman, S., Abrahams, S., Abbott, D., Mukai, Y., Samuel, M., Morrell, M., and Appels, R. (1997). A complex arrangement of genes at a starch branching enzyme I locus in the D-genome donor of wheat. Genome 40:465-474.

Rapellin, A et al 1997. Isolation of a starch branching enzyme I cDNA from a wheat endosperm library. EMBL database accession Y12320.

Russell, D.A. and Fromm, M.E. (1977). Tissue-specific expression in transgenic maize of four endosperm promoters from maize and rice. Transgenic Research 6: 157-168.

Sambrook J, Fritsch EF, Maniatis T (1989) Molecular Cloning: A Laboratory Manual,

2nd ed. Cold Spring Harbour Laboratory Press, Cold Spring Harbour, NY.

Salehuzzaman, S.N.I.M., Jacobsen, E., and Visser, R.G.F. (1992). Cloning, partial sequencing and expression of a cDNA coding for branching enzyme in cassava. Plant Mol. Biol. 20:809-819.

Sears, E.R. (1966). ***In*** **Chromosome manipulations and plant genetics** ***Edited*** **by R. Riley and K.R. Lavis, Oliver and Boyd, Edinburgh. pp29-45.**

Sheehy *et al* (1988) PNAS 85, 9905-8809.

Smith, A.M. (1988). Major differences in isoforms of starch-branching enzyme between developing embryos of round- and wrinkled-seeded peas (*Pisum sativum* L.). Planta 175:270-279.

Shure, M et al 1983. Cell 35: 225-233.

Sun, C., Sathish, P., Deiber, A., and Jansson, C. (1995). Multiple starch branching enzymes in barley endosperm. *In* Photosynthesis from light to biosphere. P. Mathis (ed.) Vol. V:317-320.

Sun, C., Sathish, P., Ahlandsberg, S., and Jansson, C. (1998). The two genes encoding starch-branching enzymes IIa and IIb are differentially expressed in Barley. Plant Physiol. 118:37-49 (earliest electronic publication date 10th September 1998).

Svegmark, K. and Hermansson, A-M. (1990). Shear induced changes in the viscoelastic behaviour of heat treated potato starch dispersions. Carboyd. Polym. 13, 29-45.

Takeda, Y., Gaun, H.P., and Preiss, J. (1993). Branching of amylose by the branching isoenzymes of maize endosperm. Carbohydrate Res. 240:252-263.

Van der Krol *et al*, Mol Gen. Genet. 220, 204-212.

Claims

1. A nucleotide sequence encoding substantially the amino acid sequence shown in Figure 10 (SEQ ID No: 2) or a functional equivalent of said nucleotide sequence.

2. A nucleotide sequence comprising substantially the sequence of B2 shown in Figure 3 (SEQ ID No: 3), or a functional equivalent thereof.

3. A nucleotide sequence comprising substantially the sequence of B4 shown in Figure 3 (SEQ ID No: 4), or a functional equivalent thereof.

4. A nucleotide sequence comprising substantially the sequence of B10 shown in Figure 3 (SEQ ID No: 5), or a functional equivalent thereof.

5. A nucleotide sequence comprising substantially the sequence of B1 shown in Figure 3 (SEQ ID No: 6), or a functional equivalent thereof.

6. A nucleotide sequence encoding substantially the amino acid sequence of B6 shown in Figure 4 (SEQ ID No: 7), or a functional equivalent thereof.

7. A portion of any of the above sequences, comprising at least 500 base pairs and having at least 90% sequence homology to the corresponding portion of the sequence from which it is derived.

8. A nucleotide sequence comprising substantially the sequence shown in Figure 5 (SEQ ID No: 8), Figure 6 (SEQ ID No: 9) or Figure 7 (SEQ ID No: 10), or a functional equivalent thereof.

9. A nucleic acid construct comprising a nucleotide sequence in accordance with any of the preceding claims.

10. A construct according to claim 9, wherein the sequence is operably linked, in sense or antisense orientation, to a promoter sequence.

11. An expression vector comprising a construct according to claim 9 or 10.

12. A host cell into which has been introduced a sequence, construct or vector in accordance with anyone of the preceding claims.

13. An amino acid sequence encoded by the nucleotide sequence of anyone of claims 1 to 8.

14. A method of altering the characteristics of a plant, comprising introducing into the plant the sequence of any one of claims 1 to 11 operably linked to a suitable promoter active in the plant so as to affect expression of a gene present in the plant.

15. A method according to claim 14, wherein the sequence is linked in the antisense orientation to the promoter.

16. A method according to claim 14 or 15, wherein the plant is a wheat plant.

17. A method according to claim 14, 15 or 16, wherein the characteristic altered relates to the starch content and/or starch composition of the plant.

18. A plant or plant cell having characteristics altered by the method of any one of claims 14 to 17, or the progeny of such a plant or part of such a plant.

19. A plant, plant cell, progeny or part thereof according to claim 18, wherein the plant is a wheat plant.

20. A storage organ from a plant according to claim 18 or 19.

21. A plant, plant cell, progeny or part thereof according to any one of claims 18 to 20,

containing starch having an elevated gelatinisation onset and/or peak temperature as measured by DSC compared to starch from a similar, but unaltered, plant.

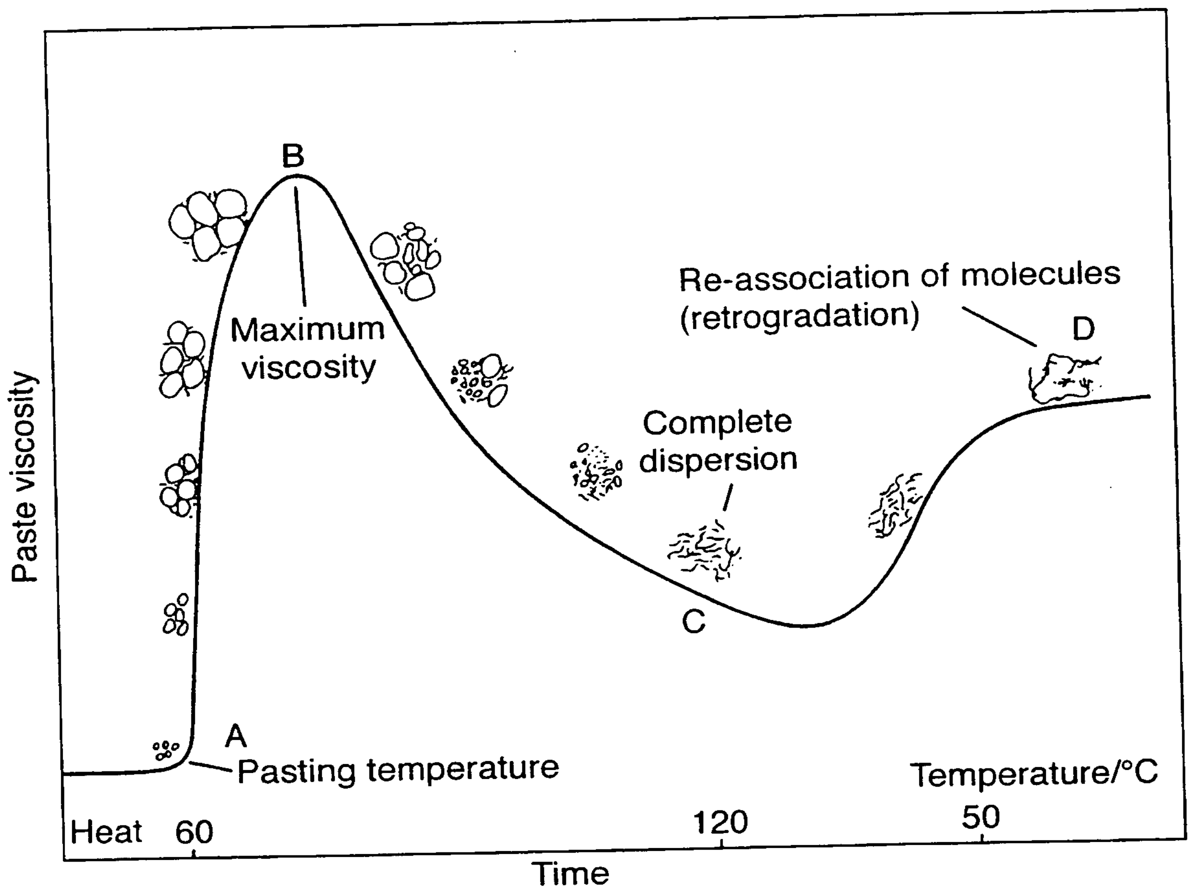
22. Starch obtainable or obtained from a plant in accordance with any one of claims 18 to 21.

23. A method of making altered starch, comprising altering a plant by the method of any one of claims 14 to 17, and extracting therefrom starch having altered properties compared to starch extracted from equivalent, but unaltered, plants.

24. Use of starch according to claim 22 in the preparation of processing of a foodstuff, particularly bakery products.

25. A foodstuff, particularly a bakery product, comprising starch in accordance with claim 22.

Fig.1.


B
Maximum viscosity
Re-association of molecules (retrogradation)
D
Complete dispersion
C
A
Pasting temperature
Temperature/°C
Paste viscosity
Heat 60
120
50
Time


OsbeII-1ALL SEQ ID No : 11
Wheat SBEII-2 SEQ ID No:12
ZMSBE2a SEQ ID No : 13
ZMSBE2b SEQ ID No : 14
Barley SBEIIa SEQ ID No : 15
Barley SBEIIb SEQ ID No : 16
RICBCE3 SEQ ID No : 17
RICESBE-1/97 SEQ ID No : 18
PSSBEIGEN SEQ ID No : 19
STSBE SEQ ID No : 20
TASBEI SEQ ID No : 21
TASBE1D2 SEQ ID No : 22
ZMSBEI SEQ ID No : 23
RICBE1 SEQ ID No : 24
PSSBEIIGN SEQ ID No : 25

Fig.2(i)

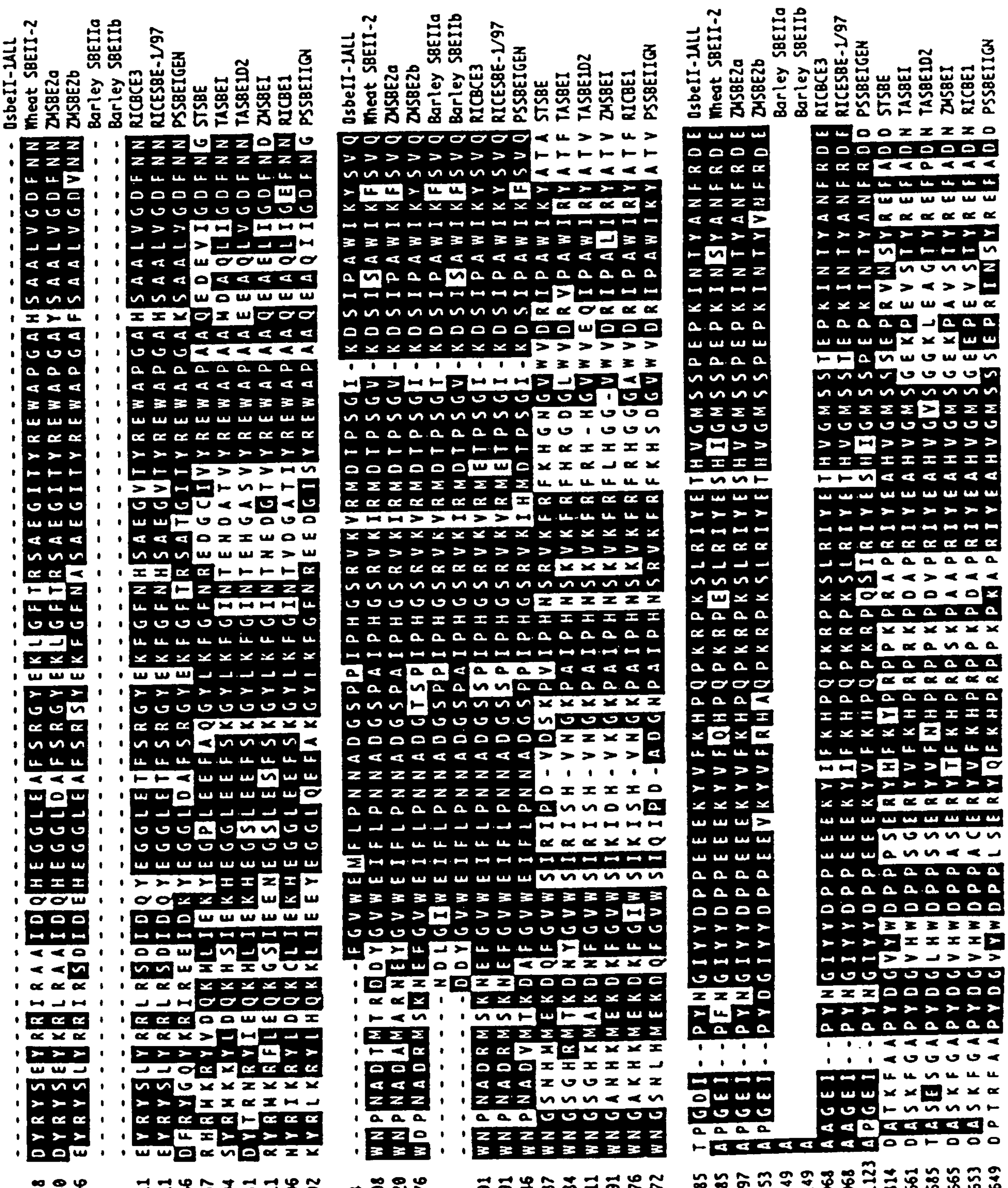

OsbeII-1ALL
Wheat SBEII-2
ZMSBE2a
ZMSBE2b
Barley SBEIIa
Barley SBEIIb
RICBCE3
RICESBE-1/97
PSSBEIGEN
STSBE
TASBEI
TASBE1D2
ZMSBEI
RICBE1
PSSBEIIGN

Fig.2(ii).

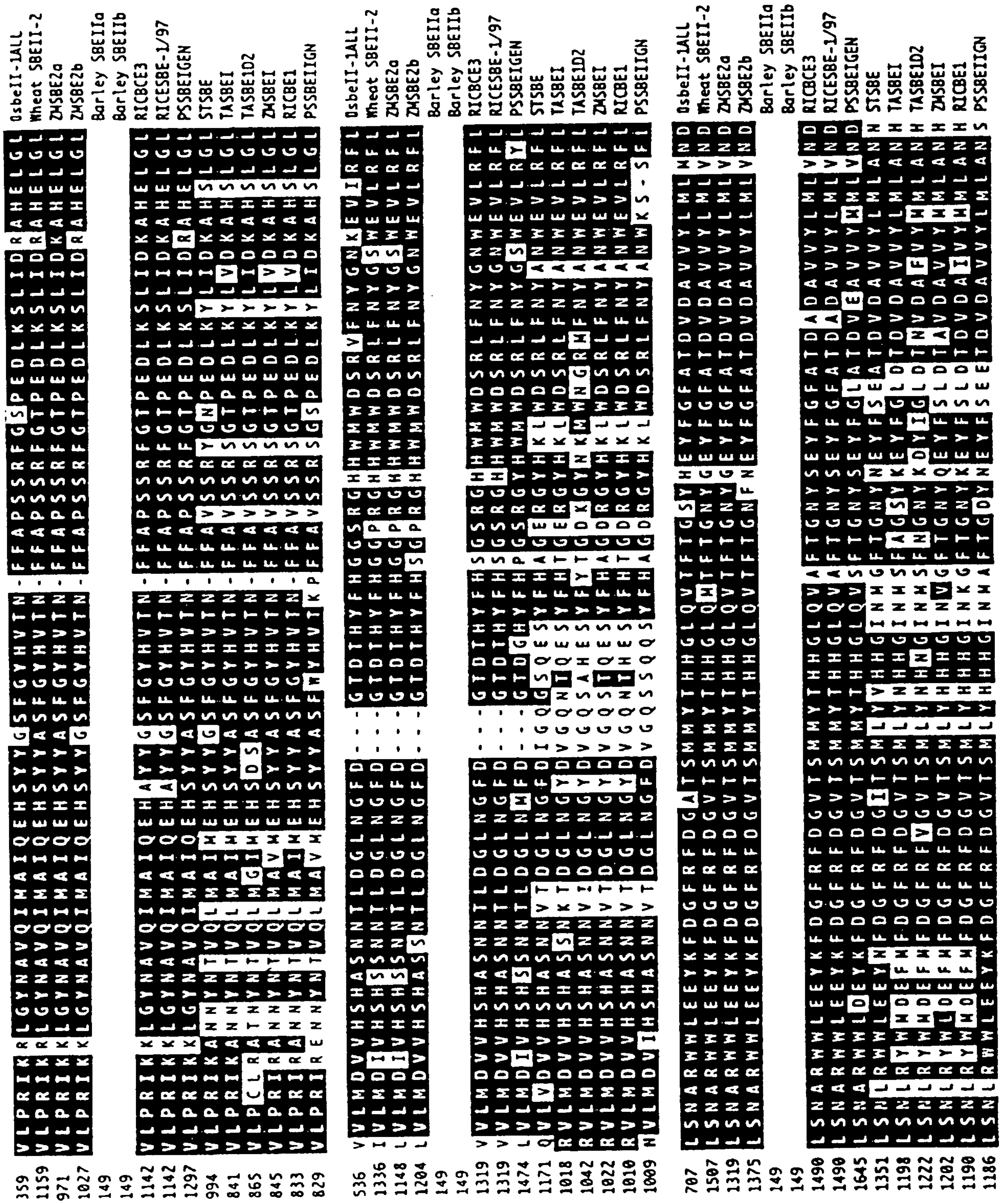

OsbeII-1ALL
Wheat SBEII-2
ZMSBE2a
ZMSBE2b
Barley SBEIIa
Barley SBEIIb
RICBCE3
RICESBE-1/97
PSSBEIGEN
STSBE
TASBEI
TASBE1D2
ZMSBEI
RICBE1
PSSBEIIGN

Fig.2(iii).

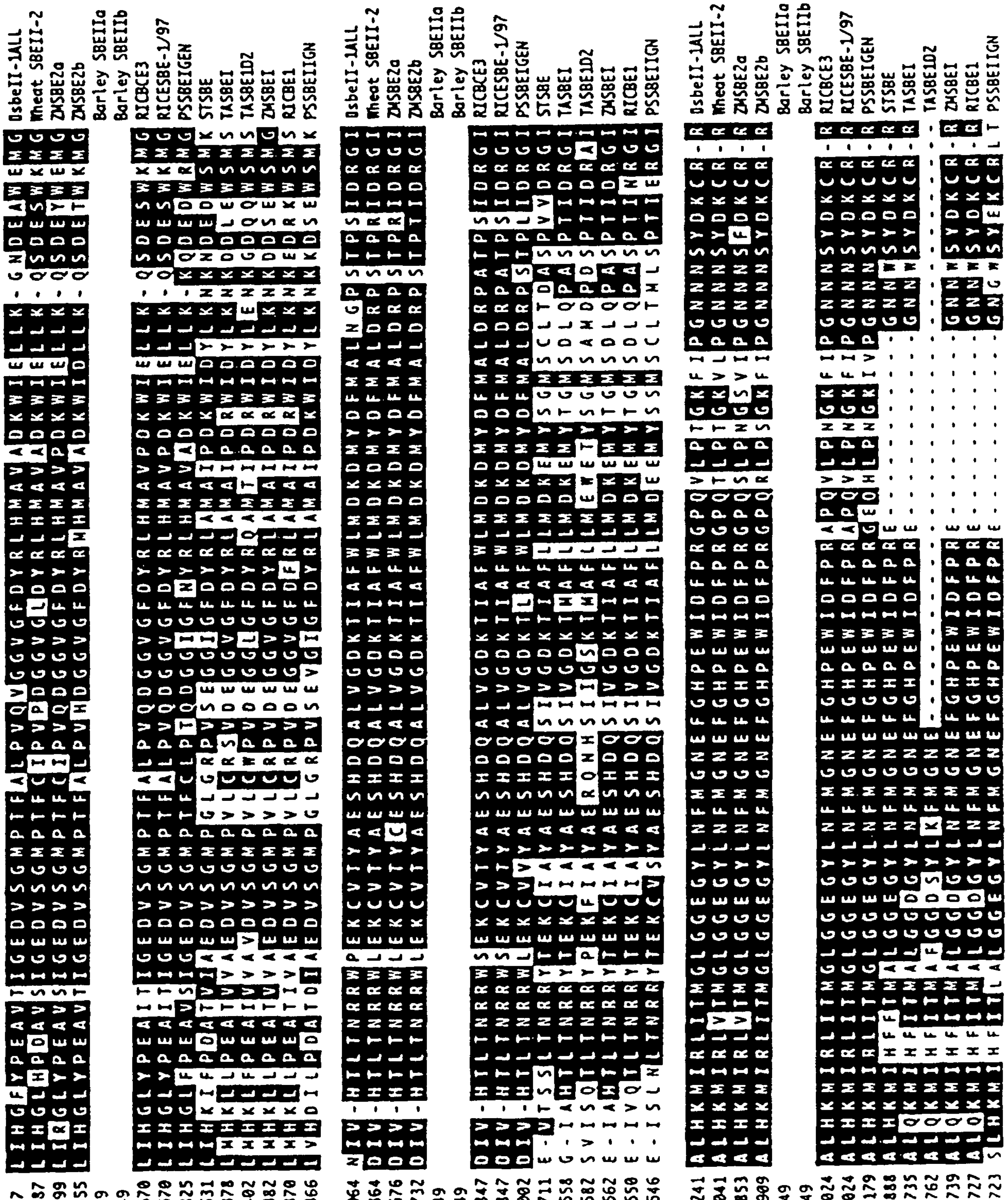


Fig.2(iv).

Fig.2(v).

Fig.2(vi).

```
1949 T--T..CALCSQIPRALWRKNAHLCYFMDQGRNLPQKPLFFL-KGG.APGICI-WMPP.I OsbeII-1ALL
2617 ------------------------------------------------------------ Wheat SBEII-2
2435 ------------------------------------------------------------ ZMSBE2a
2488                                                              ZMSBE2b
149                                                               Barley SBEIIa
149                                                               Barley SBEIIb
2585 ---------------------VYA---------------P-------------------- RICBCE3
2723 .TRQLAFV----------LCA-P-NL.F.FV------RE-RNVYL.LSMAVERRRNHEPR RICESBE-1/97
2875 PI-------------------------ERSV------EEVESETTQQSVEVESETTQQSV PSSBEIGEN
2452 LMN--ACQKL------KITRQTFVVS----------YYQQPISRRVTRNLKIRYLQISVT STSBE
2269 PSR--TC-----------------VA----------YYR--VEEKAEKPKDEGAASWGKT TASBEI
2122 Y------------------------------------PYM-GVGIKVDDIRH-I.----- TASBE1D2
2273 PPR--TC-----------------VA----------YYR--VDEAGAGRRLHAKAETGKT ZMSBEI
2261 PPR--TC-----------------VA----------YYR--VDEDREELRRGGAVASGKI RICBE1
2260 PPH--TC-----------------VV----------YYR--VDERQEESNNPNLGSVEET PSSBEIIGN

2117 FV-AINH-C.CPIN.QFRIEVIL-LYFIF-DS.TVFLK.S-TCCLLEDEK.NQRLKKKKK OsbeII-1ALL
2617 -----------------------------------------E                   Wheat SBEII-2
2435 --------E-------------------------------DE                   ZMSBE2a
2488                                                              ZMSBE2b
149                                                               Barley SBEIIa
149                                                               Barley SBEIIb
2597 --------A--------------------------------E                   RICBCE3
2846 IFVG--TV-R------------TASYT.FC----------TSVHLVHLES           RICESBE-1/97
2962 EVES--ETT-------------------------------Q.                   PSSBEIGEN
2578 LTNACQKLKFTRQTFLVSYYQQPILRRVTR------KLKDSLSTNI-----------STY STSBE
2356 ALG-YIDVE-ATGVKDAADGEATSGSEKAS------TGGDSSKKGINFVFLSPDKD-NN  TASBEI
2173 ---------------------------------------------TMLEYGGNG--GA.. TASBE1D2
2360 SPAESIDVK-AS------RASSKED----K------EATAGGKKGWKFARQPSDQDTKY  ZMSBEI
2348 V-TEYIDVE-AT------SGETISGGWKGS------EKDDCGKKGMKFVFRSSDEDCKY  RICBE1
2347 FAAADTDVARIPDVSMES--EDSNLDRIED------NSEDAVDAGILKVEREVVGDN.   PSSBEIIGN

2282 KKKKKKKKN                                                    OsbeII-1ALL
2620                                                              Wheat SBEII-2
2444                                                              ZMSBE2a
2488                                                              ZMSBE2b
149                                                               Barley SBEIIa
149                                                               Barley SBEIIb
2603                                                              RICBCE3
2918                                                              RICESBE-1/97
2986                                                              PSSBEIGEN
2704                                                              STSBE
2503                                                              TASBEI
2212 KKN                                                          TASBE1D2
2483                                                              ZMSBEI
2480                                                              RICBE1
2494                                                              PSSBEIIGN
```

Fig.2A.

Percent Similarity

Percent Divergence

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15		
1		67.9	68.8	71.4	85.7	81.6	71.4	72.5	66.8	46.6	45.4	30.4	45.5	45.5	44.4	1	sbell-1ALL
2	14.9		84.3	80.6	85.7	100.0	79.2	78.1	77.6	48.5	49.9	36.7	50.0	49.9	48.0	2	Wheat SBEII-2
3	13.9	14.6		81.0	87.8	93.9	81.7	78.1	75.9	47.1	49.5	37.5	49.9	49.7	48.1	3	ZMSBE2a
4	10.5	22.2	21.3		85.7	79.6	86.1	86.1	75.9	49.4	50.9	36.5	50.5	50.6	49.0	4	ZMSBE2b
5	11.5	15.9	13.4	15.9		85.7	85.7	85.7	85.7	32.7	26.5	30.6	30.6	28.6	36.7	5	Barley SBEIIa
6	16.6	0.0	6.4	23.9	15.9		79.6	79.6	87.8	36.7	32.7	32.7	32.7	28.6	42.9	6	Barley SBEIIb
7	10.3	23.5	22.7	14.3	15.9	23.9		100.0	75.8	50.0	50.5	37.5	51.2	50.7	49.1	7	RICBCE3
8	20.8	26.3	26.0	14.3	15.9	23.9	0.1		67.9	49.9	51.0	37.9	51.9	51.3	49.5	8	RICESBE-1/97
9	29.3	24.5	26.6	27.4	15.9	13.4	28.7	39.5		47.9	49.1	37.2	50.0	50.0	48.1	9	PSSBEIGEN
10	66.2	57.7	60.3	58.1	91.7	79.9	56.0	65.5	67.4		68.3	49.0	71.1	70.0	72.6	10	STSBE
11	68.4	58.6	59.3	58.2	121.4	98.3	57.1	66.1	67.5	38.2		58.7	82.6	83.3	67.9	11	TASBEI
12	88.4	88.7	89.9	84.9	118.1	95.3	85.1	93.8	96.7	58.8	38.0		57.2	58.5	46.7	12	TASBE1D2
13	66.6	60.0	61.1	59.6	127.2	102.3	57.8	65.7	67.9	33.8	19.1	41.1		85.2	71.4	13	ZMSBEI
14	67.8	59.8	60.9	59.2	105.4	105.4	58.0	67.7	67.2	36.4	16.6	38.2	14.9		70.1	14	RICBE1
15	65.7	60.0	61.1	59.3	79.9	64.6	57.2	66.6	68.5	28.8	38.9	61.0	33.1	34.9		15	PSSBEIIGN
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15		

Fig.3(i).

```
                                        10        20        30        40        50
     ATATGTATGATTTCATGGCTCTGGATGGACCTTCGACTCCTCGTATTGAT Majority SEQ ID No:53
  1  AT--GTATGATTTCATGGCTCTGAACGGACCTTCGACGCCTAATATTGAT B2.seq  SEQ ID No:3
  1  AT--GTATGATTTCATGGCTCTGAACGGACCTTCGACACCTAATATTGAT B4.seq  SEQ ID No:4
  1  AT--GTATGATTTCATGGCGCTGAACGGACCTTCGACGCCTAATATTGAT B10.seq SEQ ID No:5
  1  ATATGTATGATTTCATGGCTCTGGATAGACCTTCAACTCCTCGCATTGAT A2.seq  SEQ ID No:26
  1  ATATGTATGATTTCATGGCTCTGGATAGGCCTTCAACTCCTCGCATTGAT B1.seq  SEQ ID No:6
  1  ATATGTATGATTTCATGGCTCTGGATAGACCTTCAACTCCTCGCATTGAT B11.seq SEQ ID No:27

                                        60        70        80        90       100
     CGTGGCATAGCATTGCATAAAATGATTAGGCTTGTCACCATGGGTTTAGG Majority
 49  CGTGGAATAGCACTGCATAAAATGATTANACTTATCACAATGGGTTTAGG B2.seq
 49  CGTGGAATAGCACTGCATAAAATGATTAGACTTATCACAATGGGTTTAGG B4.seq
 49  CGTGGAATAGCACTGCATAAAATGATTAGACTTATCACAATGGGTCTAGG B10.seq
 51  CGTGGCATAGCATTACATAAAATGATCAGGCTTGTCACCATGGGTTTAGG A2.seq
 51  CGTGGCATAGCATTACATAAAATGATCAGGCTTGTCACCATGGGTTTAGG B1.seq
 51  CGTGGCATAGCATTACATAAAATGATCAGGCTTGTCACCATGGGTTTAGG B11.seq

                                       110       120       130       140       150
     TGGAGAGGGTTATCTTAACTTTATGGGAAATGAGTTTGGGCATCCTGAAT Majority
 99  CGGAGAGGGTTATCTTAACTTTATGGGAAATGAGTTCGGGCATCCTGAAT B2.seq
 99  AGGAGAGGGTTATCTTAACTTTATGGGAAATGAGTTCGGGCATCCTGAAT B4.seq
 99  AGGAGAGGGTTATCTTAACTTTATGGGAAATGAGTTCGGGCATCCTGAAT B10.seq
101  TGGCGAAGGCTATCTTAACTTCATGGGAAATGAGTTTGGGCATCCTGAAT A2.seq
101  TGGTGAAGGCTATCTTAACTTCATGGGAAATGAGTTTGGGCATCCTGAAT B1.seq
101  TGGCGAAGGCTATCTTAACTTCATGGGAAATGAGTTTGGGCATCCTGAAT B11.seq

                                       160       170       180       190       200
     GGATAGATTTTCCAAGAGGCCCACAAGTTCTTCCAACTGGTAAGTTTCTC Majority
149  GGATAGACTTTCCAAGAGGCCCACAAGTACTTCCAAGTGGTAAGTTCATC B2.seq
149  GGATAGACTTTCCAAGAGGCCCACAAGTACTTCCAACTGGTAAGTTCATC B4.seq
149  GGATAGACTTTCCAAGAGGCCCACAAGTACTTCCAAGTGGTAAGTTCATC B10.seq
151  GGATAGATTTTCCAAGAGGTCCGCAAACTCTTCCAACCGGCAAAGTTCTC A2.seq
151  GGATAGATTTTCCAAGAGGCCCACAAACTCTTCCAACCGGCAAAGTTCTC B1.seq
151  GGATAGATTTTCCAAGAGGTCCGCAAACTCTTCCAACCGGCAAAGTTCTC B11.seq
```

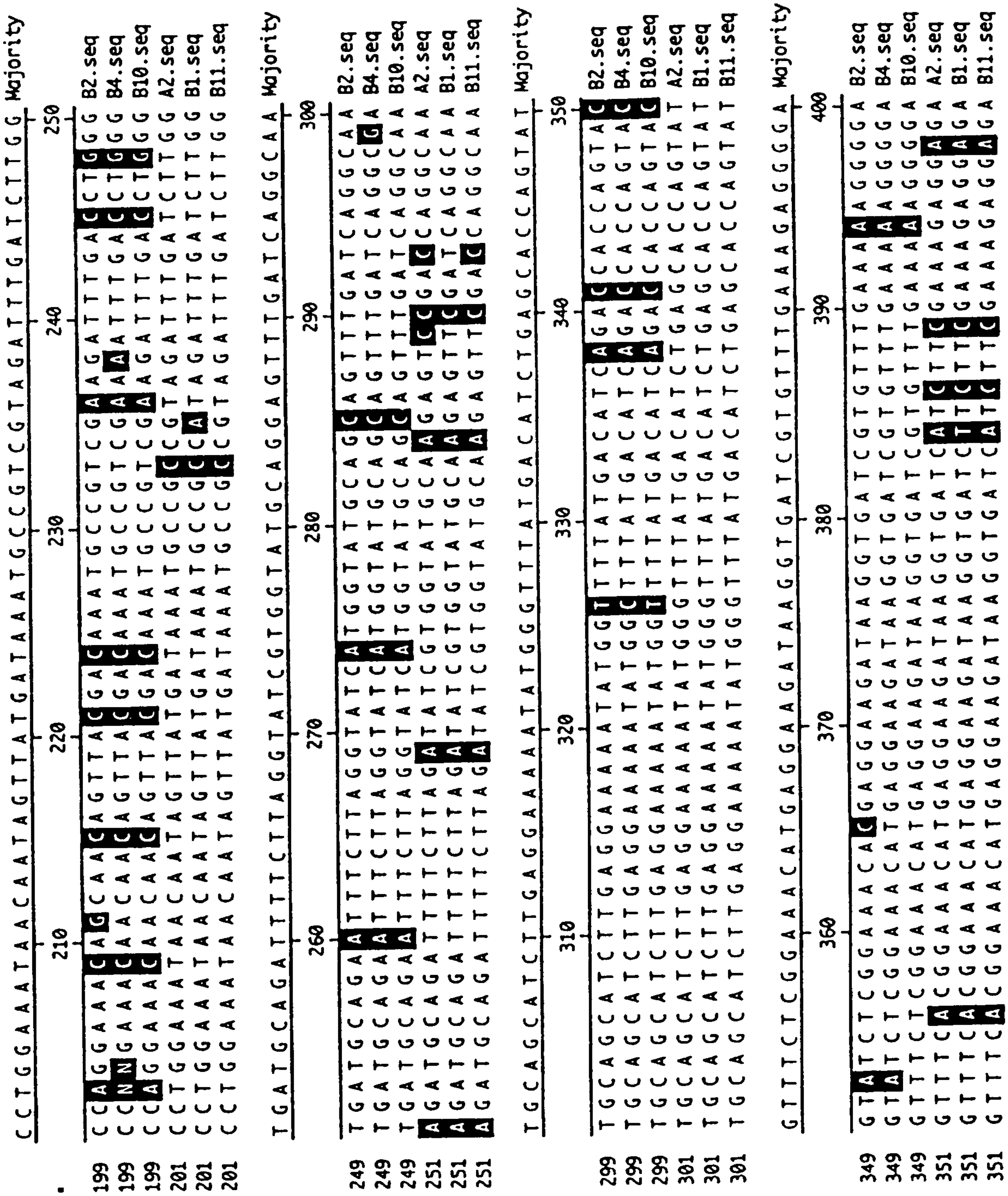


Fig.3(ii).

Fig.3(iii).

```
    TTTGGTATTTGTTTTCAACTTCCACTGGAGTAATAGCTTTTTTGACTACC Majority
             410       420       430       440       450
399 CTTGGTATTTGTGTTCAACTTCCACTGGAGTAATAGCTATTTCGACTACC B2.seq
399 CTTGGTATTTGTGTTCAACTTCCACTGGAGTAATAGCTATTTCGGCTACC B4.seq
399 CTTGGTATTTGTGTTCAACTTCCACTGGAGTAGTAGCTATTTCGACTACC B10.seq
401 TTTGGTATTCGTTTTCAACTTCCACCGGAGCAATAGCTTTTTTGACTACC A2.seq
401 TTTGGTATTTGTTTTCAACTTCCACTGGAGCAATAGCTTTTTTGACTACC B1.seq
401 TTTGGTATTTGTTTTCAACTTCCACTGGAGCAATAGCTTTTTTGACTACC B11.seq

    GTGTTGGGTGTTTCAAGCCTGGGAAGTACAAGGTGGTCTTAGACTCCGAC Majority
             460       470       480       490       500
449 GGGTCGGCTGTTTAAAGCCTGGGAAGTACAAGGTGGTCTTAGACTCAGAC B2.seq
449 GGGTTGGCTGTTTAAAGCCTGGGAAGTACAAGGTTGTCTTAGACTCAGAC B4.seq
449 GGGTCGGCTGTTTAAAGCCTGGGAAGTACAAGGTGGTCTTAGACTCGGAC B10.seq
451 GTGTTGGGTGTTCCAGGCCTGGGAAGTACAAGGTGGCCTTAGACTCCGAC A2.seq
451 GTGTTGGGTGTTCCAAGCCTGGGAAGTACAAGGTGGCCTTGGACTCCGAC B1.seq
451 GTGTTGGGTGTTCCAAGCCTGGGAAGTACAAGGTGGCCTTAGACTCCGAC B11.seq

    GCTGGACTCTTTGGTGGATTTGGTAGGCTTGATCATGCTGTCGAGTACTT Majority
             510       520       530       540       550
499 GCTGGACTCTTTGGTGGATTTGGTAGGATCCATCACACTGCAGAGCACTT B2.seq
499 GCCGGACTCTTTGGTGGATTTGGTAGGATCCATCACACTGCAGAGCACTT B4.seq
499 GCTGGACTCTTTGGTGGATTTGGTAGGATCCATCACACTGCAGAGCACTT B10.seq
501 GATGCACTCTTTGGTGGATTCAGCAGGCTTGATCATGATGTCGACTACTT A2.seq
501 GATGCACTCTTTGGTGGATTCAGCAGGCTTGATCATGATGTCGACTACTT B1.seq
501 GATGCACTCTTTGGTGGATTCAGCAGGCTTGATCATGATGTCGACTACTT B11.seq

    CACTTCTGACTGTCCGCATGACAACAGGCCGCATTCTTTCTCGGTGTACA Majority
             560       570       580       590       600
549 CACTTCTGACTGCCAACATGACAACAGGCCCCATTCGTTCTCAGTGTACA B2.seq
549 CACTTCTGACTGCCAACATGACAACAGGCCCCATTCGTTCTCAGTGTACA B4.seq
549 CACTTCTGACTGCCAACATGACAACAGGCCCCATTCATTCTCAGTGTACA B10.seq
551 CACAACCGAACATCCGCATGACAACAGGCCGCGCTCTTTCTCGGTGTACA A2.seq
551 CACAACCGAACATCCGCATGACAACAGGCCGCACTCTTTCTCGGTGTACA B1.seq
551 CACAACCGAACATCCGCATGACAATAGGCCGCGCTCTTTCTTGGTGTACA B11.seq
```

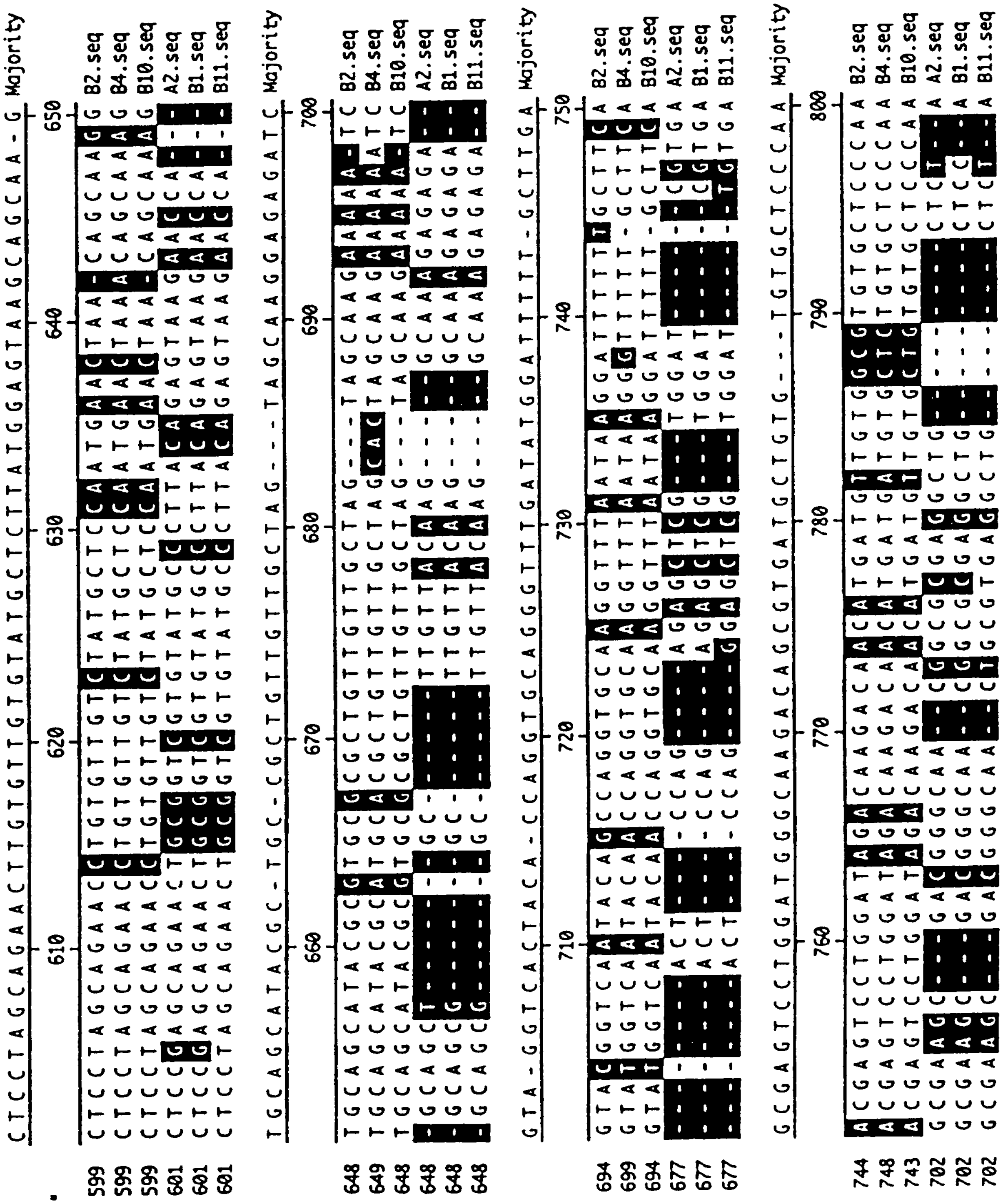


Fig.3(iv).

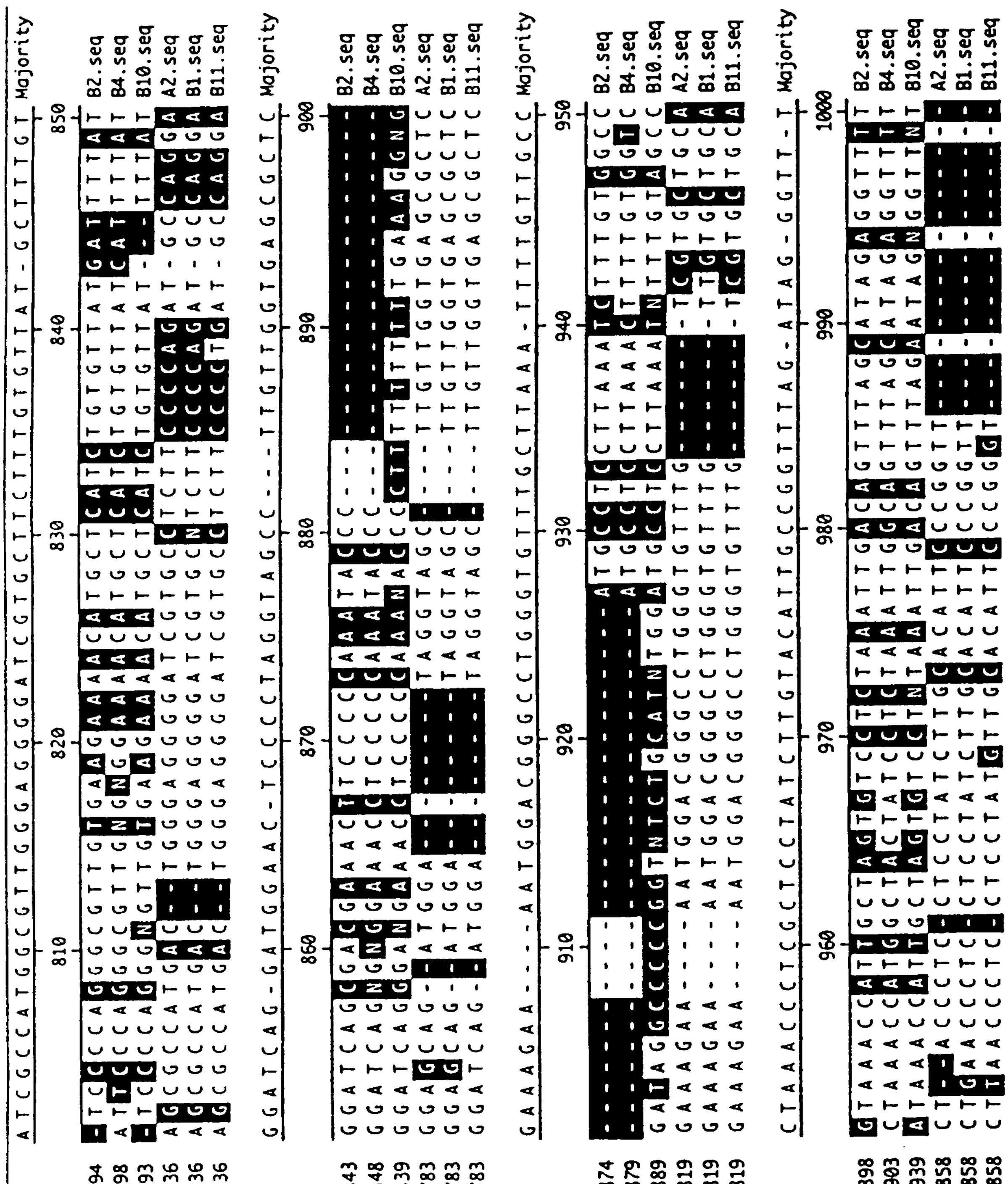


Fig.3(v).

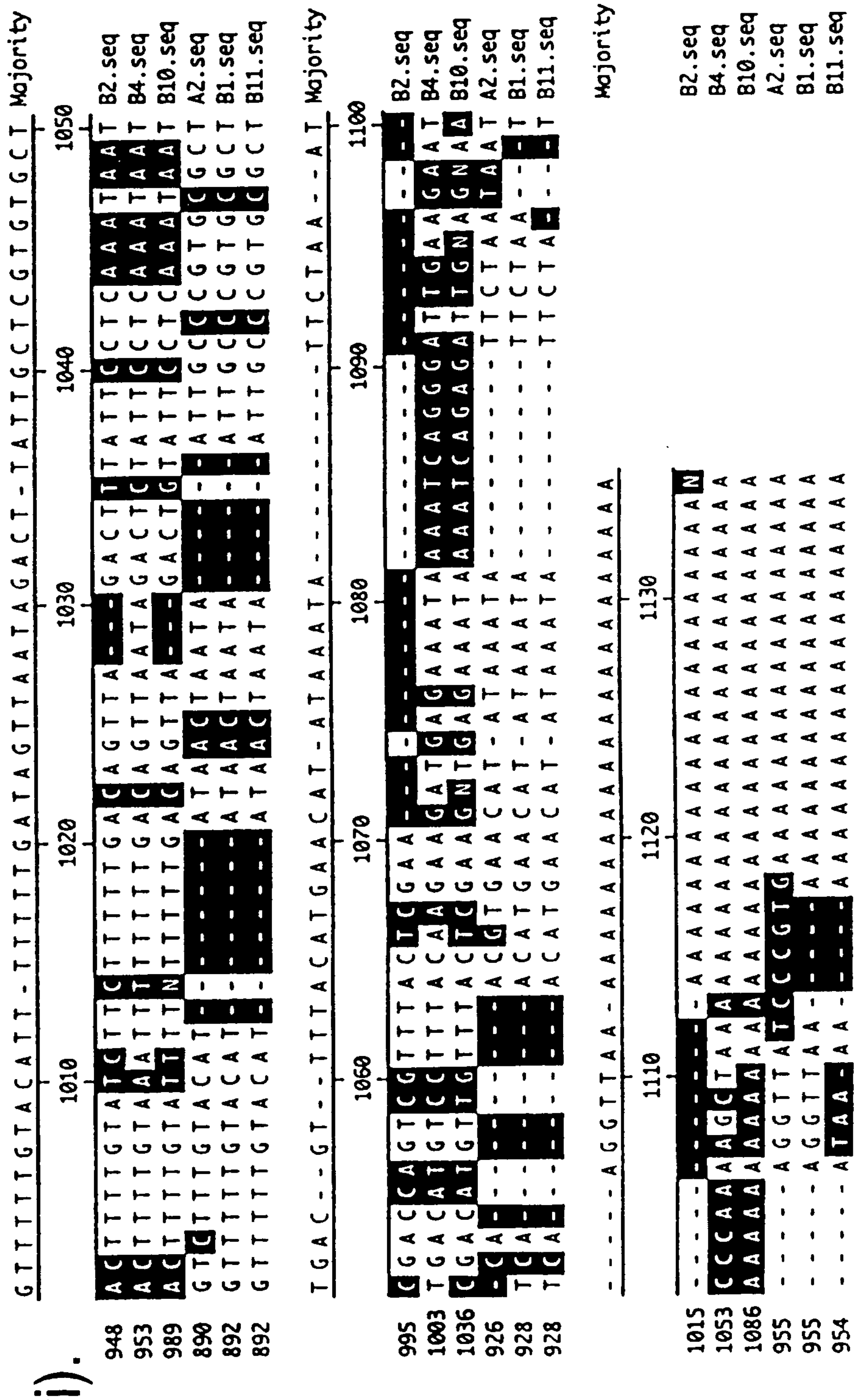


Fig.3(vi).

Decoration 'Decoration #1': Shade (with solid black) residues that differ from the Consensus.

Fig.3A.

Percent Similarity

Percent Divergence

	1	2	3	4	5	6		
1		91.0	94.4	59.0	60.0	59.5	1	B2.seq
2	4.5		89.2	58.8	59.9	59.6	2	B4.seq
3	2.4	4.6		59.3	59.6	59.8	3	B10.seq
4	32.6	32.3	34.3		95.5	95.7	4	A2.seq
5	30.5	29.7	32.0	2.1		96.8	5	B1.seq
6	31.6	30.9	32.6	2.4	2.7		6	B11.seq
	1	2	3	4	5	6		

Fig.4A.

Percent Similarity

Percent Divergence

	1	2	3	4		
1		88.7	81.7	85.0	1	Maizellb.pro
2	10.8		82.2	82.6	2	B6.pro
3	17.9	17.5		86.9	3	B11.pro
4	14.6	17.0	12.7		4	Maizella.pro
	1	2	3	4		

Fig.4.

```
1    M Y D F M A L D R P S T P T I D R G I A L H K M I R L I T M  MaizeIIb.pro SEQ ID No : 30
1    M Y D F M A L N G P S T P N I D R G I A L H K M I R L I T M  B6.pro SEQ ID No : 7
1    M Y D F M A L D R P S T P R I D R G I A L H K M I R L V T M  B11.pro SEQ ID No : 28
1    M Y D F M A L D R P S T P R I D R G I A L H K M I R L V T M  MaizeIIa.pro SEQ ID No : 29

31   G L G G E G Y L N F M G N E F G H P E W I D F P R G P Q R L  MaizeIIb.pro
31   G L G G E G Y L N F M G N E F G H P E W I D F P R G P Q V L  B6.pro
31   G L G G E G Y L N F M G N E F G H P E W I D F P R G P Q T L  B11.pro
31   G L G G E G Y L N F M G N E F G H P E W I D F P R G P Q S L  MaizeIIa.pro

61   P S G K F I P G N N N S Y D K C R R R F D L G D A D Y L R Y  MaizeIIb.pro
61   P S G K F I P G N S N S Y D K C R R R F D L G D A E F L R Y  B6.pro
61   P T G K V L P G N N N S Y D K C R R R F D L G D A D F L R Y  B11.pro
61   P N G S V I P G N N N S F D K C R R R F D L G D A D Y L R Y  MaizeIIa.pro

91   H G M Q E F D Q A M Q H L E Q K Y E F M T S D H Q Y I S R K  MaizeIIb.pro
91   H G M Q Q F D Q A M Q H L E E K Y G F M T S D H Q Y V S R K  B6.pro
91   R G M Q E F D Q A M Q H L E E K Y G F M T S E H Q Y V S R K  B11.pro
91   R G M Q E F D Q A M Q H L E G K Y E F M T S D H S Y F S R K  MaizeIIa.pro

121  H E E D K V I V F E K G D L V F V F N F H C N N S Y F D Y R  MaizeIIb.pro
121  H E E D K V I V F E K G D L V F V F N F H W S N S Y F D Y R  B6.pro
121  H E E D K V I I F E R G D L V F V F N F H W S N S F F D Y R  B11.pro
121  H E E D K V I I F E R G D L V F V F N F H W S N S Y F D Y R  MaizeIIa.pro

151  I G C R K P G V Y K V V L D S D A G L F G G F S R I H H A A  MaizeIIb.pro
151  V G C L K P G K Y K V V L D S D A G L F G G F G R I H H T A  B6.pro
151  V G C S K P G K Y K V A L D S D D A L F G G F S R L D H D V  B11.pro
151  V G C F K P G K Y K I V L D S D D G L F G G F S R L D H D A  MaizeIIa.pro

181  E H F T A D C S H D N R P Y S F S V Y T P S R T C V V Y A P  MaizeIIb.pro
181  E H F T S D C Q H D N R P H S F S V Y T P S R T C V V Y A P  B6.pro
181  D Y F T T E H P H D N R P R S F L V Y T P S R T A V V Y A L  B11.pro
181  E Y F T A D W P H D N R P C S F S V Y A P S R T A V V Y A P  MaizeIIa.pro

211  V - - - E .                                                  MaizeIIb.pro
211  M - - - N .                                                  B6.pro
211  T - - - E .                                                  B11.pro
211  A G A E D E                                                  MaizeIIa.pro
```

Decoration 'Decoration #1': Shade (with solid black) residues that differ from MaizeIIb.pro.

Fig.5.

```
        10         20         30         40         50         60
ACTAACAGCA AGGTGCAGCA TACGCGTGCG CGCTGTTGTT GCTAGTAGCA AGAAAAATCG 60
TACGGTCAAT ACAGCCAGGT GCAAGGTTTA ATAAGGATTT TTTGCTTCAA CGAGTCCTGG 120
ATAGACAAGA CAACATGATG TTGTGGCGTG TGCTCCCAAT CCCCAGGGCG TTGTGAAGAA 180
AACATGCTCA TCTGTGTTAT GATTTTATGG ATCAGCGACG AAACTTCCCC CAAATACCCA 240
TGCCTCCTTA AATCTTTGTG GCCGTAAACC ATTGCTAGTG TCCTCTAAAT TGACAGTTTA 300
       310        320        330        340        350        360
GCATAGAGGT TTTACTTTTG TATCTTCTTT TTGACAGTTA GACTTTATTC CTCAAATAAT 360
CGACCAGTCG TTTACTCG 378    (SEQ ID No : 8)
```

Fig.6.

```
        10         20         30         40         50         60
AACTAACAGC AAAGTGCAGC ATACGCGTGC GCGCTGTTGT TGCTAGTAGC AAGAAAAATC 60
GTATGGTCAA TACAACCAGG TGCAAGGTTT AATAAGGATT TTTGCTTCAA CGAGTCCTGG 120
ATAGACAAGA CAACATGATG TTGTGCTGTG TGCTCCCAAT CCCCAGGGXG TTGTGAAGAA 180
AACATGCTCA TCTGTGTTAT TTTATGGATC AGGGAXGAAA CCTCCCCCAA AXACCCCTTT 240
TTTTTTTGAA AGGXGGATAG GCCCCCGGTX TCTGCATXTG GATGCCTCCT TAAATXTTTG 300

       310        320        330        340        350        360
TAGCCATAAA CCATTGCTAG TGTCCTXTAA ATTGACAGTT TAGAATAGXG GTTXTACTTT 360
TGTATTTTXT TTTTGACAGT TAGACTGTAT TCCTCAAATA ATCGACATGT TGTTTACTCG 420
AAGXTGAGAA ATAAAATCAG AGATTGXAG 449     (SEQ ID N0 : 9)
```

Fig.7.

```
        10         20         30         40         50         60
ACTAAACAGC AAAGTGCAGC ATACGCATGC ACGCTGTTGT TGCTAGCACT AGCAAGAAAA 60
AATCGTATGG TCAATACAAC CAGGTGCAAG GTTTAATAAG GGTTTTTGCT TCAACGAGTC 120
CTGGATAGAC AAGACAACAT GATGATGTGC TCTGTGCTCC CAAATTCCCA GGGCGTTGXG 180
XGGAAAACAT GCTCATCTGT GTTATCATTT TATGGATCAG XGXGGAAACC TCCCCCAAAT 240
ACCCATGCCT CCTTAAACTT TTGTGGTCCT AAACCATGGC TACTATCCTC TAAATTGGCA 300
       310        320        330        340        350        360
GTTTAGCATA GAGGTTTTAC TTTTGTAAAT TTTTTTTGAC AGTTAATAGA CTCTATTCCT 360
CAAATAATTG ACATGTCCTT TACAAGAAGA TGAGAAATAA AATCAGGGAT TGAAGAATCC 420
CAAAAGCT 428 (SEQ ID No : 10)
```

Fig.8(i).

```
1   A A C T A A C A G C A A A G T G C A G C A T A C G C G T G C  B10-3'.seq SEQ ID No : 9
1   A - C T A A C A G C A A G G T G C A G C A T A C G C G T G C  B2-3'.seq SEQ ID No : 8
1   A C T A A A C A G C A A A G T G C A G C A T A C G C A T G C  B4-3'.seq SEQ ID No : 10
1   - - - - - - T A G C G G G G T A C - - - - - - - - - - - - -  ZMSBE2b-3'.seq SEQ ID No : 31

31  G C G C T G T T G T T G C T A G - - - T A G C A A G A A A A  B10-3'.seq
30  G C G C T G T T G T T G C T A G - - - T A G C A A G A A A A  B2-3'.seq
31  A C G C T G T T G T T G C T A G C A C T A G C A A G A A A A  B4-3'.seq
12  - - - - - - T C G T T G C T - G C G C - G G C A - - - - - -  ZMSBE2b-3'.seq

58  A - T C G T A T G G T C A A T A C A A C C A G G T G C A A G  B10-3'.seq
57  A - T C G T A C G G T C A A T A C A G C C A G G T G C A A G  B2-3'.seq
61  A A T C G T A T G G T C A A T A C A A C C A G G T G C A A G  B4-3'.seq
28  - - - T G T G T G G - - - - G G C T G T C - G A T G T G A G  ZMSBE2b-3'.seq

87  G T T T A A T A A G G A T T T T T - G C T T C A A C G A G T  B10-3'.seq
86  G T T T A A T A A G G A T T T T T T G C T T C A A C G A G T  B2-3'.seq
91  G T T T A A T A A G G G T T T T T - G C T T C A A C G A G T  B4-3'.seq
50  G - - - - - - A A A A A C C T T C T - - T C C A A - - A A C  ZMSBE2b-3'.seq

116 C C T G G A T A G A C A A G A C A A C A T G A T G T T G T G  B10-3'.seq
116 C C T G G A T A G A C A A G A C A A C A T G A T G T T G T G  B2-3'.seq
120 C C T G G A T A G A C A A G A C A A C A T G A T G A T G T G  B4-3'.seq
70  C - - - G G C A G A T G - - - - - - C A T G - - - - C A T G  ZMSBE2b-3'.seq

146 C T G T G T G C T C C C A A - T C C C C A G G G N G T T G T  B10-3'.seq
146 G C G T G T G C T C C C A A - T C C C C A G G G C G T T G T  B2-3'.seq
150 C T C T G T G C T C C C A A A T T C C C A G G G C G T T G N  B4-3'.seq
87  C - - - A T G C T A C - - A A T - - - - A A G G T - - - - -  ZMSBE2b-3'.seq

175 G A A G A A A A C A T G C T C A T C T G T G T T A T - - - T  B10-3'.seq
175 G A A G A A A A C A T G C T C A T C T G T G T T A T G A T T  B2-3'.seq
180 G N G G A A A A C A T G C T C A T C T G T G T T A T C A T T  B4-3'.seq
103 - - - - - - - - - - - - - - - - T C T G - - - - A T - A C T  ZMSBE2b-3'.seq

202 T T A T G G A T C A G G G A N G A A A C C T C C C C C A A A  B10-3'.seq
205 T T A T G G A T C A G C G A C G A A A C T T C C C C C A A A  B2-3'.seq
210 T T A T G G A T C A G N G N G G A A A C C T C C C C C A A A  B4-3'.seq
112 T T A - - - A T C G - - - - - - - - - - - - - - - - - - - A  ZMSBE2b-3'.seq
```

Fig.8(ii).

```
232 N A C C C C T T T T T T T T T T G A A A G G N G G A T A G G B10-3'.seq
235 T A C C C - - - - - - - - - - - - - - - - - - - - - - - - - B2-3'.seq
240 T A C C C - - - - - - - - - - - - - - - - - - - - - - - - - B4-3'.seq
120 T G C - - - - - - - - - - - - T G G A A A G C C C A T G C A ZMSBE2b-3'.seq

262 C C C C C G G T N T C T G C A T N T G G A T G C C T C C T T B10-3'.seq
240 - - - - - - - - - - - - - - - - - - - - A T G C C T C C T T B2-3'.seq
245 - - - - - - - - - - - - - - - - - - - - A T G C C T C C T T B4-3'.seq
138 T C - - - - - T C G C T G C G T T - - - G T - C C T C T C T ZMSBE2b-3'.seq

292 A A A T N T T T G T A G C C A T A A A C C A T T G C T A G T B10-3'.seq
250 A A A T C T T T G T G G C C G T A A A C C A T T G C T A G T B2-3'.seq
255 A A A C T T T T G T G G T C C T A A A C C A T G G C T A C T B4-3'.seq
159 A - - - - - - T A T A - - T A T A A - - - - - - - - - - - - ZMSBE2b-3'.seq

322 G T C C T N T A A A T T G A C A G T T T A G A A T A G N G G B10-3'.seq
280 G T C C T C T A A A T T G A C A G T T T A G C A T A G A G G B2-3'.seq
285 A T C C T C T A A A T T G G C A G T T T A G C A T A G A G G B4-3'.seq
169 G A C C T T C A A G G T G T C A A T T A A A C A T A G A G T ZMSBE2b-3'.seq

352 T T N T A C T T T T G T A T T T T N T T T T T G A C A G T T B10-3'.seq
310 T T T T A C T T T T G T A T C T T C T T T T T G A C A G T T B2-3'.seq
315 T T T T A C T T T T G T A A A T T T T T T T T G A C A G T T B4-3'.seq
199 T T T C G T T T T T - - - - - - - - - - - - - - - - - - - - ZMSBE2b-3'.seq

382 A - - - G A C T G T A T T C C T C A A A T A A T C G A C A T B10-3'.seq
340 A - - - G A C T T T A T T C C T C A A A T A A T C G A C C A B2-3'.seq
345 A A T A G A C T C T A T T C C T C A A A T A A T T G A C A T B4-3'.seq
209 - - - - - - - - - - - - - - - - - - - - - - - - - - - - - - ZMSBE2b-3'.seq
```

Fig.8(iii).

```
409 G T T G T T T A C T C G A A G N T G A G A A A T A A A A T C B10-3'.seq
367 G T C G T T T A C T C G                                     B2-3'.seq
375 G T C C T T T A C A A G A A G A T G A G A A A T A A A A T C B4-3'.seq
209 - - C G C T T T C - - - - - - - - - - - - - - - - - - - - - ZMSBE2b-3'.seq

439 A G A G A T T G N A G                                       B10-3'.seq
378                                                             B2-3'.seq
405 A G G G A T T G A A G A A T C C C A A A A G C T             B4-3'.seq
216 - - - - - - - - - - - - - - - - - - - - - - C T             ZMSBE2b-3'.seq
```

Decoration 'Decoration #1': Shade (with solid black) residues that differ from B10-3'.seq.

Fig.8A.

Percent Similarity

Percent Divergence	1	2	3	4		
1		88.9	76.2	26.3	1	B10-3'.seq
2	4.1		81.2	31.8	2	B2-3'.seq
3	7.2	9.4		29.5	3	B4-3'.seq
4	33.5	32.6	33.9		4	ZMSBE2b-3'.seq
	1	2	3	4		

Fig.9A.

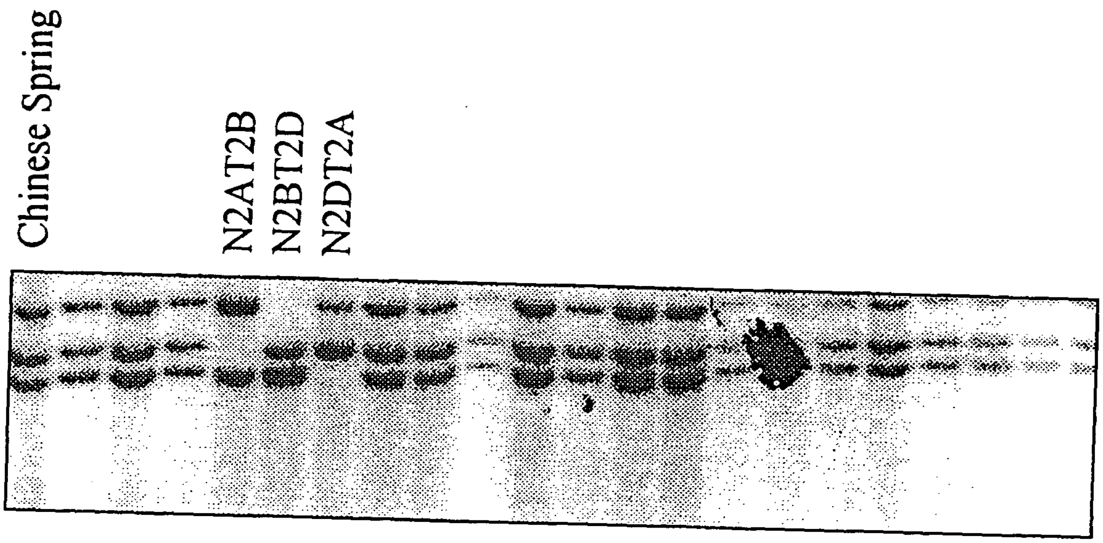


Fig.9B.

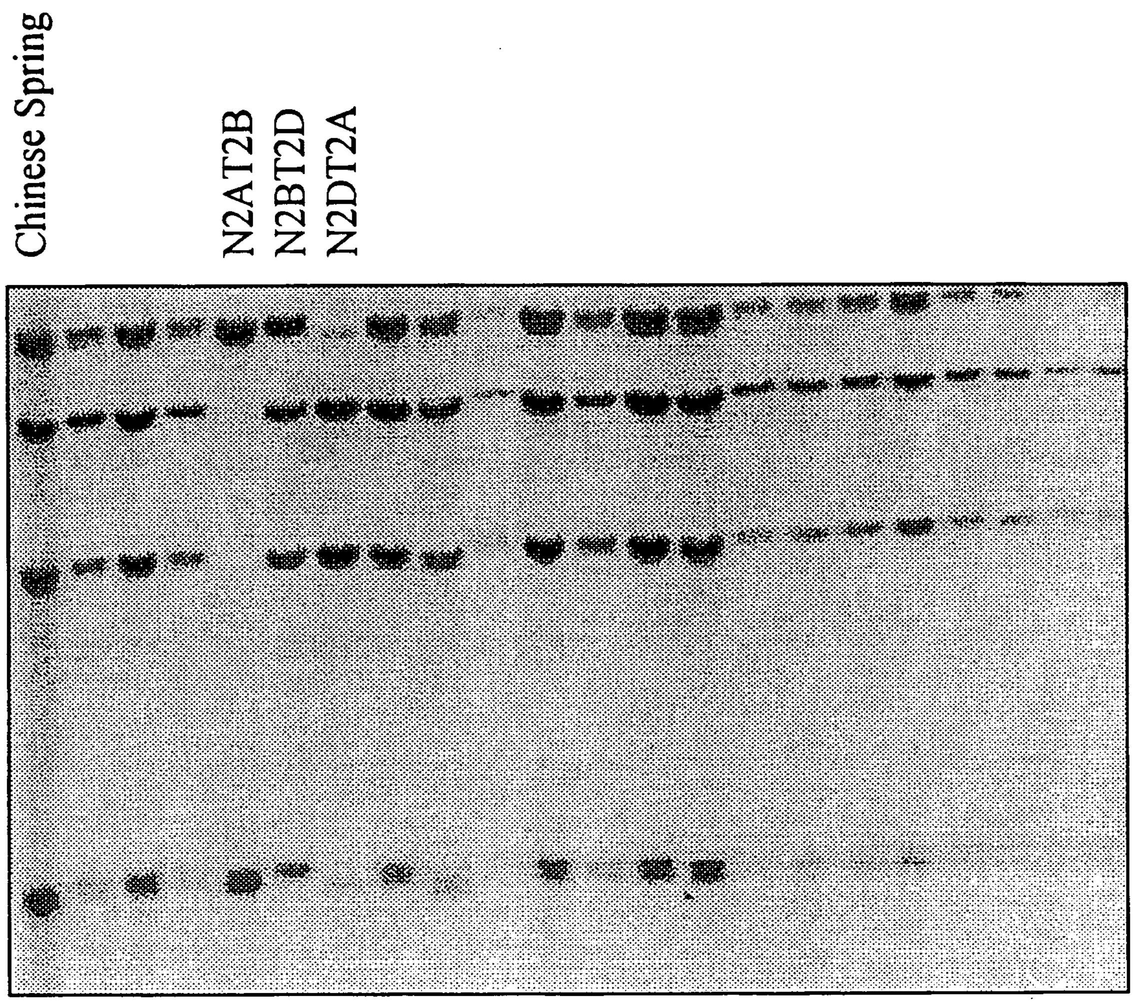


Fig.10(i).

```
CATYGACGGCCAGTGACTTCGAGCTCGGTACCCGGGGATCCGATTTGGTGTGTGGGAGATGTTCTTGCCAAACAATGCAGATGGTTCGCC 90 SEQ ID No : 1
  I  D  G  Q  .  L  R  A  R  Y  P  G  I  R  F  G  V  W  E  M  F  L  P  N  N  A  D  G  S  P    SEQ ID No : 2

ACCAATTCCTCACGGCTCACGGGTGAAGGTGAGAATGGATACTCCATCTGGGATAAAGGATTCAATTCCTGCTTGGATCAAGTACTCCGT 180
  P  I  P  H  G  S  R  V  K  V  R  M  D  T  P  S  G  I  K  D  S  I  P  A  W  I  K  Y  S  V

GCAGACTCCAGGAGATATACCATACAATGGAATATATTATGATCCTCCCGAAGAGGAGAAGTATGTATTCAAGCATCCTCAACCTAAACG 270
  Q  T  P  G  D  I  P  Y  N  G  I  Y  Y  D  P  P  E  E  E  K  Y  V  F  K  H  P  Q  P  K  R

ACCAAAATCATTGCGGATATATGAAACACATGTTGGCATGAGTAGCCCGGAACCAAAGATCAACACATATGCAAACTTCAGGGATGAGGT 360
  P  K  S  L  R  I  Y  E  T  H  V  G  M  S  S  P  E  P  K  I  N  T  Y  A  N  F  R  D  E  V

GCTTCCAAGAATTAAAAGACTTGGATACAATGCAGTGCAAATAATGGCAATCCAGGAGCACTCATACTATGGAAGCTTTGGGTACCATGT 450
  L  P  R  I  K  R  L  G  Y  N  A  V  Q  I  M  A  I  Q  E  H  S  Y  Y  G  S  F  G  Y  H  V

TACCAATTTCTTTGCACCAAGTAGCCGTTTTGGGTCCCCAGAAGATTTAAAATCTTTGATTGATAGAGCTCACGAGCTTGGCTTGGTTGT 540
  T  N  F  F  A  P  S  S  R  F  G  S  P  E  D  L  K  S  L  I  D  R  A  H  E  L  G  L  V  V

CCTCATGGATGTTGTTCACAGTCACGCGTCAAATAATACCTTGGACGGGTTGAATGGTTTTGATGGCACGGATACACATTACTTCCATGG 630
  L  M  D  V  V  H  S  H  A  S  N  N  T  L  D  G  L  N  G  F  D  G  T  D  T  H  Y  F  H  G

CGGTTCACGGGGCCATCACTGGATGTGGGATTCCCGTGTGTTTAACTATGGGAATAAGGAAGTTATAAGGTTTCTACTTTCCAATGCAAG 720
  G  S  R  G  H  H  W  M  W  D  S  R  V  F  N  Y  G  N  K  E  V  I  R  F  L  L  S  N  A  R

ATGGTGGCTAGAGGAGTATAAGTTTGATGGTTTCCGATTCGATGGCGCGACCTCCATGATGTATACCCATCATGGATTACAAGTAACCTT 810
  W  W  L  E  E  Y  K  F  D  G  F  R  F  D  G  A  T  S  M  M  Y  T  H  H  G  L  Q  V  T  F
```

Fig.10(ii).

```
TACAGGAAGCTACCATGAATATTTTGGCTTTGCCACTGATGTAGATGCGGTCGTTTACTTGATGCTGATGAATGATCTAATTCATGGGTT 900
 T  G  S  Y  H  E  Y  F  G  F  A  T  D  V  D  A  V  V  Y  L  M  L  M  N  D  L  I  H  G  F
TTATCCTGAAGCCGTAACTATCGGTGAAGATGTTAGTGGAATGCCTACATTTGCCCTTCCTGTTCAAGTTGGTGGGGTTGGTTTTGACTA 990
 Y  P  E  A  V  T  I  G  E  D  V  S  G  M  P  T  F  A  L  P  V  Q  V  G  G  V  G  F  D  Y
TCGCTTACATATGGCTGTTGCCGACAAATGGATTGAACTTCTCAAAGGAAACGATGAAGCTTGGGAGATGGGTAATATTGTGCACACACT 1080
 R  L  H  M  A  V  A  D  K  W  I  E  L  L  K  G  N  D  E  A  W  E  M  G  N  I  V  H  T  L
AACAAACAGAAGGTGGCCGGAAAAGTGTGTTACTTATGCTGAAAGTCACGATCAAGCACTGGTTGGAGACAAGACTATTGCATTCTGGTT 1170
 T  N  R  R  W  P  E  K  C  V  T  Y  A  E  S  H  D  Q  A  L  V  G  D  K  T  I  A  F  W  L
GATGGACAAGGATATGTATGATTTCATGGCTCTGAACGGACCTTCGACACCTAGTATTGATCGTGGAATAGCACTGCATAAAATGATTAG 1260
 M  D  K  D  M  Y  D  F  M  A  L  N  G  P  S  T  P  S  I  D  R  G  I  A  L  H  K  M  I  R
ACTTATCACAATGGGTTTAGGAGGAGAGGGTTATCTTAACTTTATGGGAAATGAGTTCGGGCATCCTGAATGGATAGACTTTCCAAGAGG 1350
 L  I  T  M  G  L  G  G  E  G  Y  L  N  F  M  G  N  E  F  G  H  P  E  W  I  D  F  P  R  G
CCCACAAGTACTTCCAACTGGTAAGTTCATCCCAGGAAACAACAACAGTTACGACAAATGCCGTCGAAGATTTGACCAGGGTGATGCAGA 1440
 P  Q  V  L  P  T  G  K  F  I  P  G  N  N  N  S  Y  D  K  C  R  R  R  F  D  Q  G  D  A  E
ATTTCTTAGGTATCATGGTATGCAGCAGTTTGATCAGGCGATGCAGCATCTTGAGGAAAAATATGGCTTTATGACATCAGACCACCAGTA 1530
 F  L  R  Y  H  G  M  Q  Q  F  D  Q  A  M  Q  H  L  E  E  K  Y  G  F  M  T  S  D  H  Q  Y
CGTATCTCGGAAACATGAGGAAGATAAGGTGATCGTGTTTGAAAAAGGGGACTTGGTATTTGTGTTCAACTTCCACTGGAGTAATAGCTA 1620
 V  S  R  K  H  E  E  D  K  V  I  V  F  E  K  G  D  L  V  F  V  F  N  F  H  W  S  N  S  Y
```

Fig.10(iii).

```
TTTCGACTACCGGGTTGGCTGTTTAAAGCCTGGGAAGTACAAGGTTGTCTTAGACTCAGACGCCGGACTCTTTGGTGGATTTGGTAGGAT 1710
 F  D  Y  R  V  G  C  L  K  P  G  K  Y  K  V  V  L  D  S  D  A  G  L  F  G  G  F  G  R  I
CCATCACACTGCAGAGCACTTCACTTCTGACTGCCAACATGACAACAGGCCCCATTCGTTCTCAGTGTACACTCCTAGCAGAACCTGTGT 1800
 H  H  T  A  E  H  F  T  S  D  C  Q  H  D  N  R  P  H  S  F  S  V  Y  T  P  S  R  T  C  V
TGTCTATGCTCCAATGAACTAAACAGCAAAGTGCAGCATACGCATGCACGCTGTTGTTGCTAGCACTAGCAAGAAAAAATCGTATGGTCA 1890
 V  Y  A  P  M  N  .  T  A  K  C  S  I  R  M  H  A  V  V  A  S  T  S  K  K  K  S  Y  G  Q
ATACAACCAGGTGCAAGGTTTAATAAGGGTTTGCTTCAACGAGTCCTGGATAGACAAGACAACATGATGATGTGCTCTGTGCTCCCAAAT 1980
 Y  N  Q  V  Q  G  L  I  R  V  C  F  N  E  S  W  I  D  K  T  T  .  .  C  A  L  C  S  Q  I
TCCCAGGGCGTTGTGGAGAAAAAATGCTCATCTGTGTTATTTTATGGATCAGGGANGAAACCTCCCCCAAANACCCCTTTTTTTTTTGAA 2070
 P  R  A  L  W  R  K  N  A  H  L  C  Y  F  M  D  Q  G  ?  N  L  P  Q  ?  P  L  F  F  L  K
AGGNGGATAGGCCCCCGGTNTCTGCATNTGGATGCCTCCTTAAATNTTTGTAGCCATAAACCATTGCTAGTGTCCTNTAAATTGACAGTT 2160
 G  G  .  A  P  G  ?  C  ?  W  M  P  P  .  ?  F  V  A  I  N  H  C  .  C  P  ?  N  .  Q  F
TAGAATAGNGGTTNTACTTTTGTATTTTNTTTTTGACAGTTAGACTGTATTCCTCAAATAATCGACATGTTGTTTACTCGAAGNTGAGAA 2250
 R  I  ?  V  ?  L  L  Y  F  ?  F  D  S  .  T  V  F  L  K  .  S  T  C  C  L  L  E  ?  E  K
ATAAAATCAGAGATTGNAGNAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA 2307
 .  N  Q  R  L  ?  ?  K  K  K  K  K  K  K  K  K  K  K  N
```

Fig.11(i).

```
    MLCL---------------SXSLLPRP--------------------SRA Majority SEQ ID No:54
             10        20        30        40        50
52  MLCL---------------SSSLLPRP--------------------S-A TASBE1D2 SEQ ID No:32
16  MLCLT----------APSCSPSLPPRP--------------------SRP TASBEI SEQ ID No:33
44  -------------------------------------------------- OsbeII-1ALL SEQ ID No:11
151 MATFAVSGWTLGVARPAGAGGGLLPRSGSERRGGVDLPSLLLRKKDSSRA Wheat SBEII-2 SEQ ID No:34

    AADRPX-PGI--XGGGXXRLSAVPA-PXXLR-------------WXWPRK Majority
             60        70        80        90        100
94  AADRPL-PGIIAGGGGKRLSVVPSVPFLLR-------------WLWPRK TASBE1D2
76  AADRPG-PGI--SGGGNVRLSAVPA-PSSLR-------------WSWPRK TASBEI
44  -------------------------------------------------- OsbeII-1ALL
301 VLSRAASPGKVLVPDGESDDLASPAQPEELQIPEDIEEQTAEVNMTGGTA Wheat SBEII-2

    AKSKSSVPVXAXXXXIXATXXXGVXXLP----------------IYDLDP Majority
             110       120       130       140       150
202 AKSKSFVSVTARGNKIAATTGYGSDHLP----------------IYDLDL TASBE1D2
175 AKSKFSVPVSAPRDYTMATAEDGVGDLP----------------IYDLDP TASBEI
44  -------------------------------------------------- OsbeII-1ALL
451 EKLESSEPTQGIVETITDGVTKGVKELVVGEKPRVVPKPGDGQKIYEIDP Wheat SBEII-2

    KLAXFKXHFDYRXXXYXXQKHXIEKHEGGLEEFSKGYLKFGINTEXXAXV Majority
             160       170       180       190       200
304 KLAEFKDHFDYTRNRYIEQKHLIEKHEGSLEEFSKGYLKFGINTEHGASV TASBE1D2
277 KFAGFKEHFSYRMKKYLDQKHSIEKHEGGLEEFSKGYLKFGINTENDATV TASBEI
44  -------------------------------------------------- OsbeII-1ALL
601 TLKDFRSHLDYRYSEYRRIRAAIDQHEGGLEAFSRGYEKLGFTRSAEGIT Wheat SBEII-2

    YREWAPAAXXAQLVGDFNNWNGSGHXMTKDNFGVWSIRLSNNADGSPAIP Majority
             210       220       230       240       250
454 YREWAPAAEEAQLVGDFNNWNGSGHKMAKDNFGVWSIRISH-VNGKPAIP TASBE1D2
427 YREWAPAAMDAQLIGDFNNWNGSGHRMTKDNYGVWSIRISH-VNGKPAIP TASBEI
44  --------------------------------FGVWEMFLPNNADGSPPIP OsbeII-1ALL
751 YREWAPGAHSAALVGDFNNWNPNADTMTRDDYGVWEIFLPNNADGSPAIP Wheat SBEII-2
```

Fig.11(ii).

```
     H G S K V K F R F D T P S G V W V D S I P A W I K Y A V Q T A G E I G A P Y D G I H Y D P P S E E K  Majority
                         260                 270                 280                 290                 300
601  H N S K V K F R F R H - H G V W V E Q I P A W I R Y A T V T A S E S G A P Y D G L H W D P P S S E R  TASBE1D2
574  H N S K V K F R F H R G D G L W V D R V P A W I R Y A T F D A S K F G A P Y D G V H W D P P S G E R  TASBEI
101  H G S R V K V R M D T P S G I - K D S I P A W I K Y S V Q T P G D I - - P Y N G I Y Y D P P E E E K  OsbeII-1ALL
901  H G S R V K I R M D T P S G V - K D S I S A W I K F S V Q A P G E I - - P F N G I Y Y D P P E E E K  Wheat SBEII-2

     Y V F K H P Q P K K P D S L R I Y E A H V G M S G P E P E I N T Y A E F R D E V L P R I K A L G Y N  Majority
                         310                 320                 330                 340                 350
748  Y V F N H P R P P K P D V P R I Y E A H V G V S G G K L E A G T Y R E F P D N V L P C L R A T N Y N  TASBE1D2
724  Y V F K H P R P R K P D A P R I Y E A H V G M S G E K P E V S T Y R E F A D N V L P R I K A N N Y N  TASBEI
242  Y V F K H P Q P K R P K S L R I Y E T H V G M S S P E P K I N T Y A N F R D E V L P R I K R L G Y N  OsbeII-1ALL
1042 Y V F Q H P Q P K R P E S L R I Y E S H I G M S S P E P K I N S Y A N F R D E V L P R I K R L G Y N  Wheat SBEII-2

     A V Q L M A I Q E H S Y Y A S F G Y H V T N F F A V S S R S G T P E D L K S L I D K A H S L G L R V  Majority
                         360                 370                 380                 390                 400
898  T V Q L M G I M E H S D S A S F G Y H V T N F F A V S S R S G T P E D L K Y L I D K A H S L G L R V  TASBE1D2
874  T V Q L M A I M E H S Y Y A S F G Y H V T N F F A V S S R S G T P E D L K Y L V D K A H S L G L R V  TASBEI
392  A V Q I M A I Q E H S Y Y G S F G Y H V T N F F A P S S R F G S P E D L K S L I D R A H E L G L V V  OsbeII-1ALL
1192 A V Q I M A I Q E H S Y Y A S F G Y H V T N F F A P S S R F G T P E D L K S L I D R A H E L G L I V  Wheat SBEII-2

     L M D V V H S H A S N N T L D G L N G F D V G Q G T D T S Y F H G G X R G H H K M W D S R L F N Y G  Majority
                         410                 420                 430                 440                 450
1048 L M D V V H S H A S N N V I D G L N G Y D V G Q S A H E S Y F Y T G D K G Y N K M W N G R M F N Y A  TASBE1D2
1024 L M D V V H S H A S S N K T D G L N G Y D V G Q N T Q E S Y F H T G E R G Y H K L W D S R L F N Y A  TASBEI
542  L M D V V H S H A S N N T L D G L N G F D - - - G T D T H Y F H G G S R G H H W M W D S R V F N Y G  OsbeII-1ALL
1342 L M D I V H S H S S N N T L D G L N G F D - - - G T D T H Y F H G G P R G H H W M W D S R L F N Y G  Wheat SBEII-2

     N W E V L R F L L S N A R Y W L D E F K F D G F R F D G V T S M L Y T H H G L N M S F T G S Y K E Y  Majority
                         460                 470                 480                 490                 500
1198 N W E V L R F L L S N L R Y W M D E F M F D G F R F V G V T S M L Y N H N G I N M S F N G N Y K D Y  TASBE1D2
1174 N W E V L R F L L S N L R Y W M D E F M F D G F R F D G V T S M L Y N H H G I N M S F A G S Y K E Y  TASBEI
683  N K E V I R F L L S N A R W W L E E Y K F D G F R F D G A T S M M Y T H H G L Q V T F T G S Y H E Y  OsbeII-1ALL
1483 S W E V L R F L L S N A R W W L E E Y K F D G F R F D G V T S M M Y T H H G L Q M T F T G N Y G E Y  Wheat SBEII-2
```

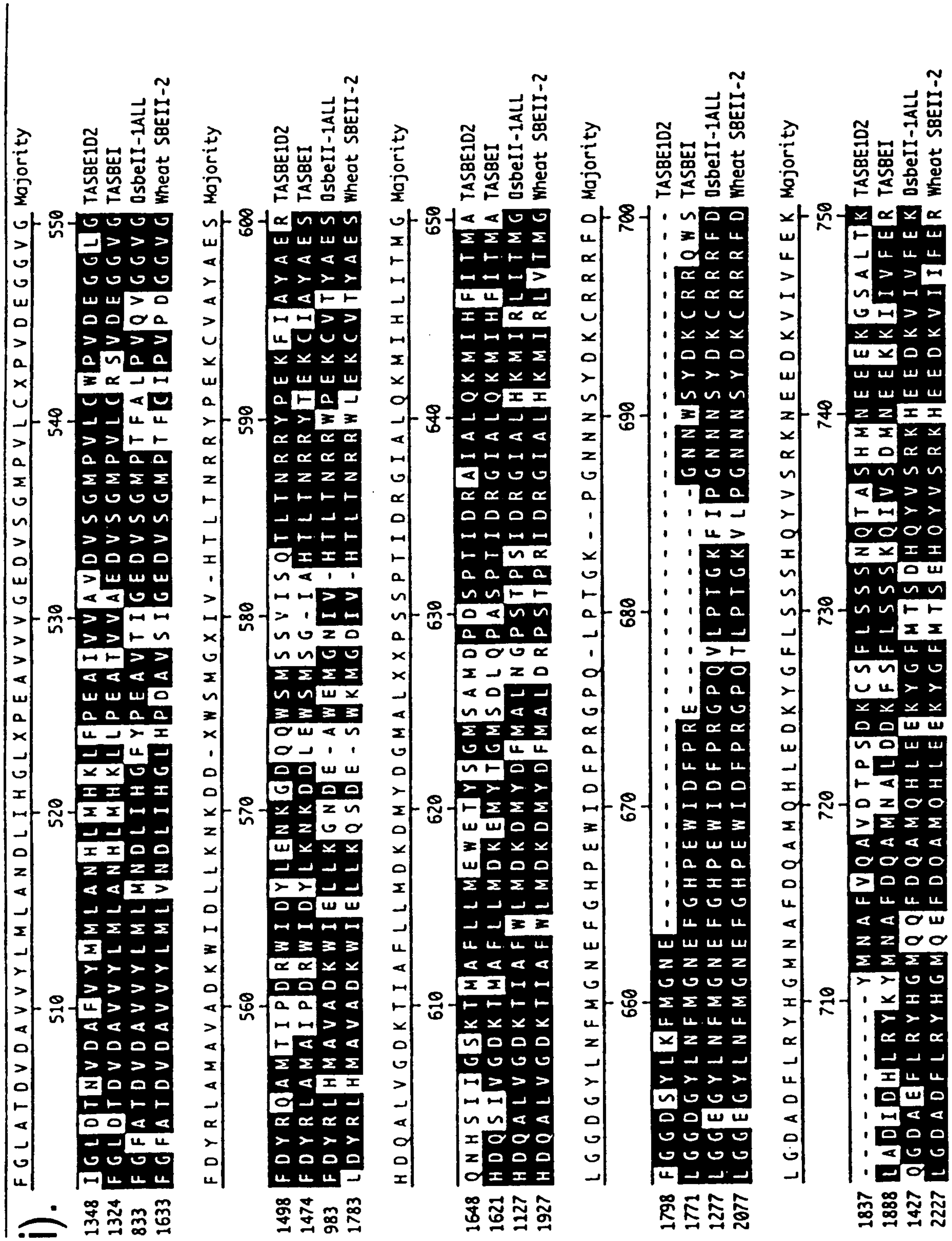


Fig.11(iii).

Fig.11(iv).

```
     GDLVFVFNFHWSNSYFDYRVGCXXPGKYKVALDSDAXLFGGFGRXXHDXD Majority
              760       770       780       790       800
1957 G-------------YTHLRSGC---------------------------- TASBE1D2
2038 GDLVFVFNFHPSKTYDGYKVGCDLPGKYKVALDSDALMFGGHGRVAHDND TASBEI
1577 GDLVFVFNFHWSNSYFDYRVGCLKPGKYKVVLDSDAGLFGGFGRIHHTAE OsbeII-1ALL
2377 GDLVFVFNFHWSNSFFDYRVGCSRPGKYKVALDSDDALFGGFSRLDHDVD Wheat SBEII-2

     HFTS--------EXXHDNRPXSFSVLTPSRTCVVY------A--P-XEXA Majority
              810       820       830       840       850
1984 -FDPS---------------------LPSTSSCA---------------- TASBE1D2
2188 HFTSPEGVPGVPETNFNNRPNSFKILSPSRTCVAYYRVEEKAEKPKDEGA TASBEI
1727 HFTS--------DCQHDNRPHSFSVYTPSRTCVVY------A--PMN-.T OsbeII-1ALL
2527 YFTT--------EHPHDNRPRSFSVYTPSRTAVVY------ALTE-.EPA Wheat SBEII-2

     AX----------V--T--K----XYXXXXXLXRXXG----XXXX--XX-- Majority
              860       870       880       890       900
2020 -------------------------------------------------- TASBE1D2
2338 ASWGKTALGYIDVEATGVKDAADGEATSGSEKASTGG---DSSKKGINFV TASBEI
1826 AKCSI--RMHAVVASTSKKKSYGQYNQVQGLIRVCFNESWIDKT--T..C OsbeII-1ALL
2632 AA----------------------CYKAKRELQRARG-S.AKRRATARG-- Wheat SBEII-2

     FLXPXKXXX-XXXXX-L-XXXXXXPXXXPXIXFXXX----GG-XXXX--- Majority
              910       920       930       940       950
2020 FLGP---SNQSPFSK-------------PFIGFPGCIFCCGL-------- TASBE1D2
2479 FLSPDKDNK-.APYQRLIRTVCRRPCNTPAIASSSNTVKLCGLEILAWTL TASBEI
1964 ALCSQIPRALWRKNAHLCYFMDQGRNLPQKPLFFL-----KGG.APG--I OsbeII-1ALL
2710 ---CSKRH----------DWEGIVPLPQ-----MPG----GA-DG.V--- Wheat SBEII-2

     X-X-XXXXKXXXXAVXX-XXXSXX-----XXXXIL--XLXXXXIIXXX-- Majority
              960       970       980       990      1000
2098 ----FKGE-.MII-----YPY---------MGVGI-----KVDDIRH-I-- TASBE1D2
2626 LRLPT-I.K-DK.AVMVRVESSYMCQICAIPS-PLS.RKFRA-SIPE.KQ TASBEI
2093 C-I-WMPP.IFV-AINH-C.CPIN-----.QFRIEV-IL-LYFIF-DS.- OsbeII-1ALL
2782 ACW.ALERKWTGLGVCRAALPSSY-----LAHSRL--FLYI.LIIARA-- Wheat SBEII-2
```

Fig.11(v).

```
     - - - - - - X X - - - X X - - - - L X - - - - - X X X - - X X X X X X X X X K K K K K K K K -   Majority
                       1010                1020                1030                1040
2170 - - - - - - - - - - - - - - - - - - . T M L E Y G G N - - G - - G - - A . . K K                 TASBE1D2
2764 L S V C K K K K K K R H E D E D A L P H R P L L L A I S P A A P - L P S R C . P A R             TASBEI
2204 - - - - - - T V F L K . S - T C C L L - - - - - E D E K . N Q R L K K K K K K K K K K K K K     OsbeII-1ALL
2905 - - - - - - L H - - - V N I - - - - - - - - - - - - - - - . I F . . . V I P . K K K K K K K     Wheat SBEII-2
```

Decoration 'Decoration #1': Shade (with solid black) residues that match the Consensus exactly.

Fig.11A.

Percent Similarity

Percent Divergence

	1	2	3	4		
1		63.9	31.2	37.0	1	TASBE1D2
2	39.1		46.7	41.8	2	TASBEI
3	86.9	73.8		69.6	3	sbeII-1ALL
4	94.5	76.4	25.3		4	Wheat SBEII-2
	1	2	3	4		

Fig.12.

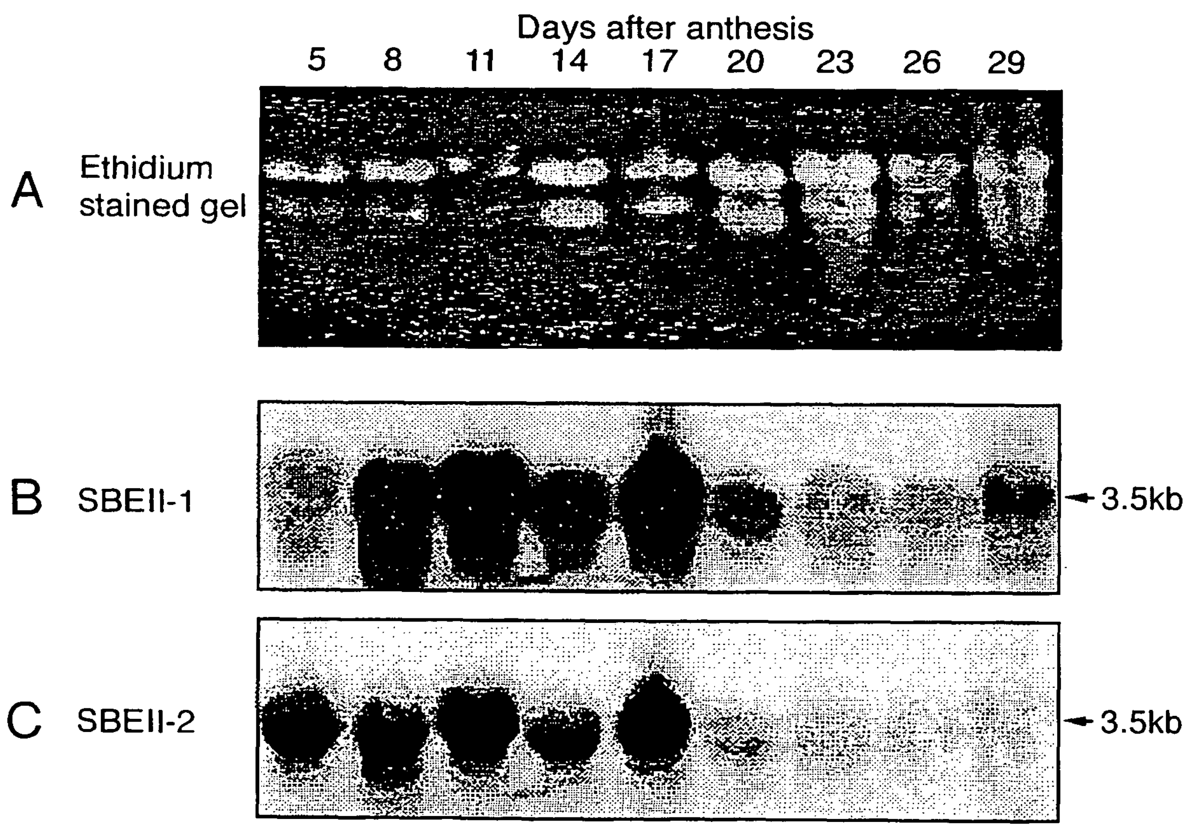


Fig.13.

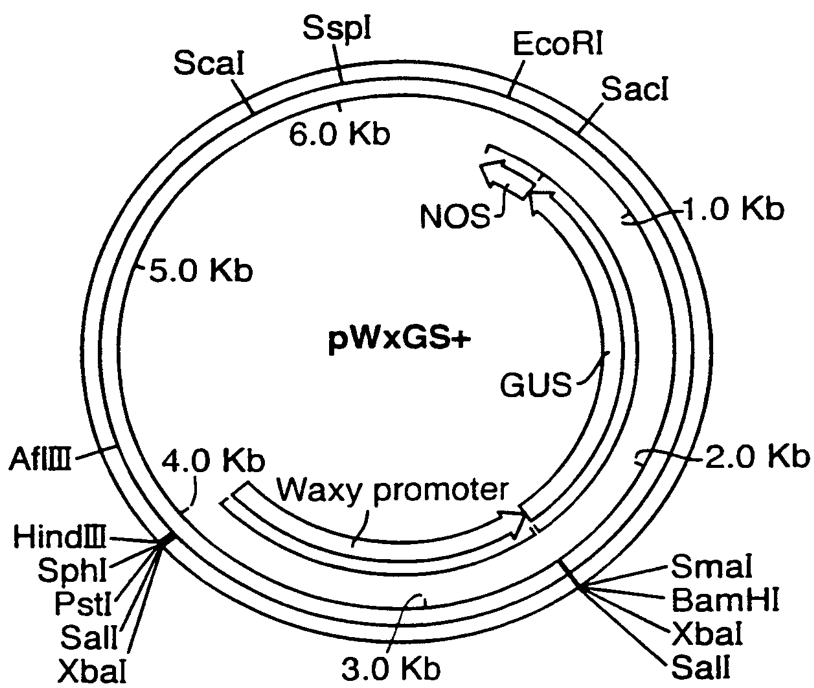

SspI
ScaI
EcoRI
SacI
6.0 Kb
NOS
1.0 Kb
5.0 Kb
pWxGS+
GUS
AflIII
4.0 Kb
2.0 Kb
Waxy promoter
HindIII
SphI
PstI
SalI
XbaI
SmaI
BamHI
XbaI
SalI
3.0 Kb

Fig.13A.

```
         10        20        30        40        50        60        70
AAGCTTGCATGCCTGCAGGTCGACTCTAGACCAAATTTCATGGTAGTTGGGAGCCTACCCAGATTTCATG 70
ATTAACTGTGCTATTGAATTGTTGAAAATGGTTGTGTCTGTCGTATCCGACGGATAACGGAAACCCGTCC 140
GAAATTCAATGGGCATGGGCATAGATATAGATTTGTACCCACTACTAGTATGGTCGCAGGCGGATATTGG 210
TTGCAACCGCAGATATAGTTTCGGGGAAAAGGATTAGGCTCAGCTCCATCCCTAGACCCCACTTGTGTGT 280
GTGGGGGGGTCTACCCTTCAAAAGGAAAAAAAACTACACACAGTGCATATAAGAAGATGAATATTCCAAA 350
        360       370       380       390       400       410       420
ATTCAGCAGTCAAGAAGCCCTGATAAACTGTCTGGCATAGCTAGTACTTTATACACTTCAAGACCAAAAG 420
AAATCACTAAGTACAGATTTTAGTGACTCGTAAGTACAGATATCATCTTACAAGGCCCAGCCCAGCGACC 490
TATTACACAGCCCGCTCGGGCCCGCGACGTCGGGACACATCTTCTTCCCCCTTTTGGTGAAGCTCTGCTC 560
GCAGCTGTCCGGCTGCTTGGACGTTCGTGTGGCAGATTCATCTGTCGTCTCGTCTCCTGTGCTTCCTGGG 630
TAGCTTGTGCAGTGGAGCTGACATGGTCTGAGCAGGCTTAAAATTTGCTCGTAGACGAGGAGTACCAGCA 700
        710       720       730       740       750       760       770
CAGCACGTTGCGGATTTCTCTGCCTGTGAAGTGCAACGTCTAGGATTGTCACACGCCTTGGTCGCGTCGA 770
TGCGGTGGTGAGCAGAGCAGCAACAGCTGGGCGGCCCAAAGTTGGCTTCCGTGTCTTCGTCGTACGTACG 840
CGCGCGCCGGGGACACGCAGAGAGCGGAGAGCGAGCCGTGCACGGGGGAGGTGGTGTGCAAGTGCAGCCG 910
CGCGCCCGCGCCCGCGCCCGGTGGGCAACCCAAAAAGTACCCACGACAAGCGAAGGCGCCAAAGCGATCC 980
AAGCTCCGGAACGCATCAGCCACAAGCAGCCGAGAACCGAACCGGTGGGCGACGCGTCGTGGGACGGACG 1050
```

Fig.13A(Cont).

```
      1060      1070      1080      1090      1100      1110      1120
CGGGCGACGCTTCCAAACGGGGCCACGTACGCCGGCGTGTGCGTGCGTGCGTGCAGACGACAAGCCAAGG 1120
CGAGGCAGCCCCCGATCGGGAAAGCGTTTTGGGCGCGAGCGCTGGCGTGCGGGTCAGTCGCTGGTGCGCA 1190
GTGCCGGGGGGAACGGGTATCGTGGGGGGCGCGGCGGAGAAGAGCGTGGCGAGGCCGAGAGCAGCGCGCG 1260
GCCGGGTCACGCAACGCGCCCCACGTACTGCCCTCCCCCTCCGCGCGCGCTAGAAATACCGAGGCCTGGA 1330
CCGGGGGCCCCCCCGTCACATCCATCCATCGACCGATCGATCGCCACAGCCAACACCACCCGCCGAGGCG 1400
      1410      1420      1430      1440      1450      1460      1470
ACGCGACAGCCGCCAGGAGGAAGGAATAAACTCACTGCCAGCCAGTGAAGGGGGAGAAGTGTACTGCTCC 1470
GTCGACTCTAGAGGATCC 1488     (SEQ ID NO:55)
```

Fig. 13.

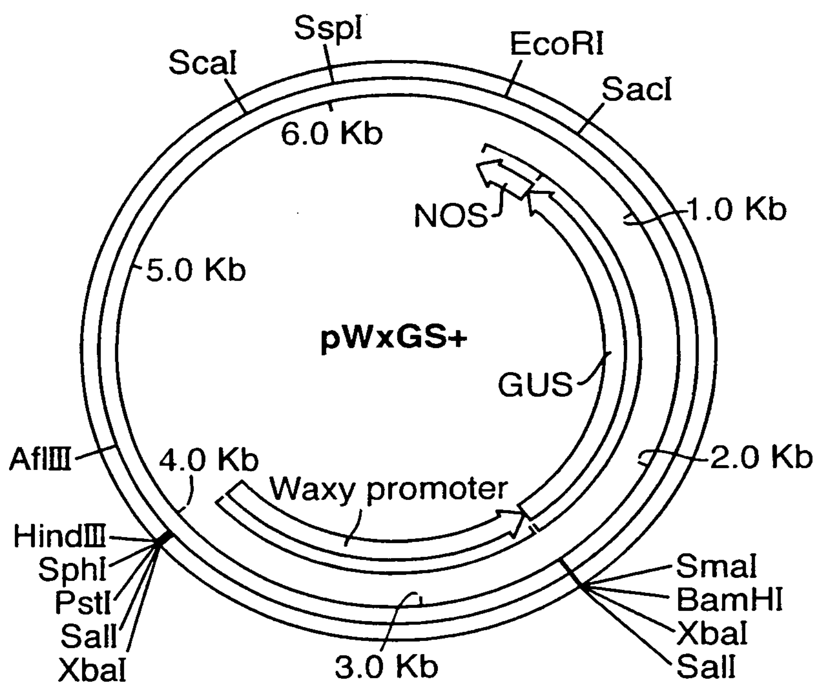

SspI
ScaI
EcoRI
SacI
6.0 Kb
NOS
1.0 Kb
5.0 Kb
pWxGS+
GUS
AflIII
4.0 Kb
2.0 Kb
Waxy promoter
HindIII
SphI
PstI
SalI
XbaI
SmaI
BamHI
XbaI
SalI
3.0 Kb

Fig.14.

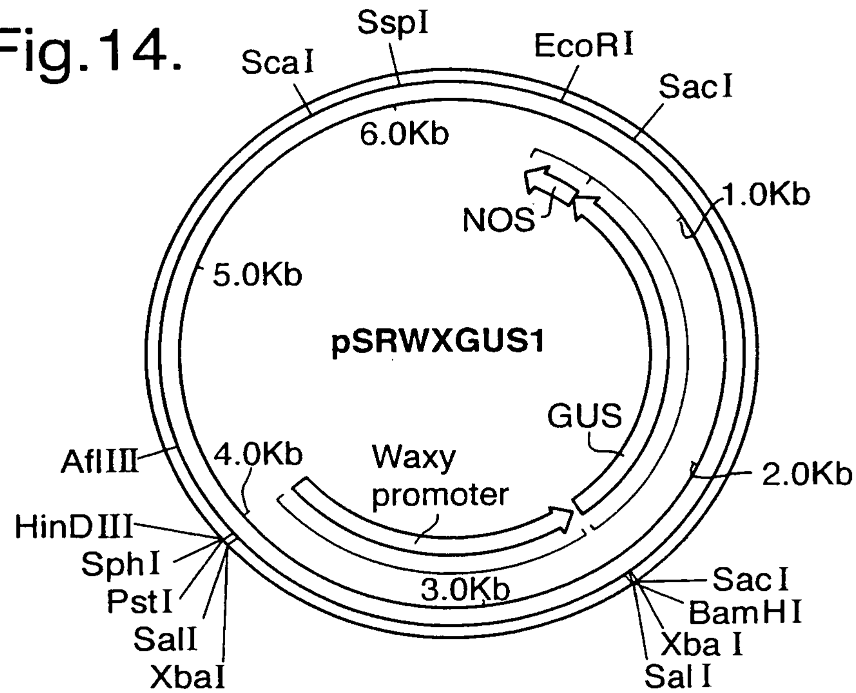

SspI
ScaI
EcoRI
SacI
6.0Kb
1.0Kb
NOS
5.0Kb
pSRWXGUS1
GUS
AflIII
4.0Kb
Waxy
promoter
2.0Kb
HinDIII
SphI
PstI
SalI
XbaI
3.0Kb
SacI
BamHI
Xba I
Sal I

Fig.15.

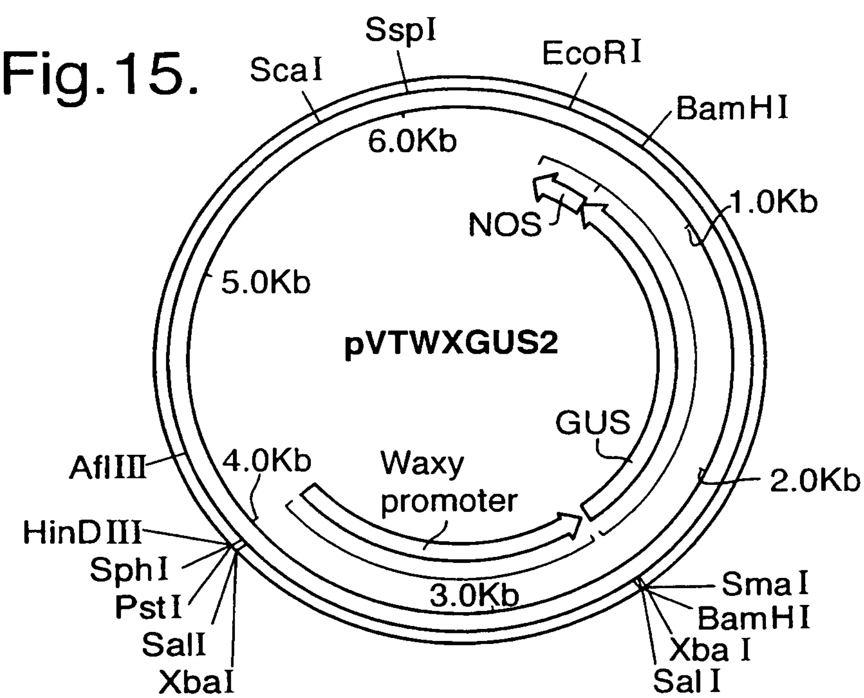

SspI
ScaI
EcoRI
BamHI
6.0Kb
1.0Kb
NOS
5.0Kb
pVTWXGUS2
GUS
AflIII
4.0Kb
Waxy
promoter
2.0Kb
HinDIII
SphI
PstI
SalI
XbaI
3.0Kb
SmaI
BamHI
Xba I
Sal I

Fig.16.

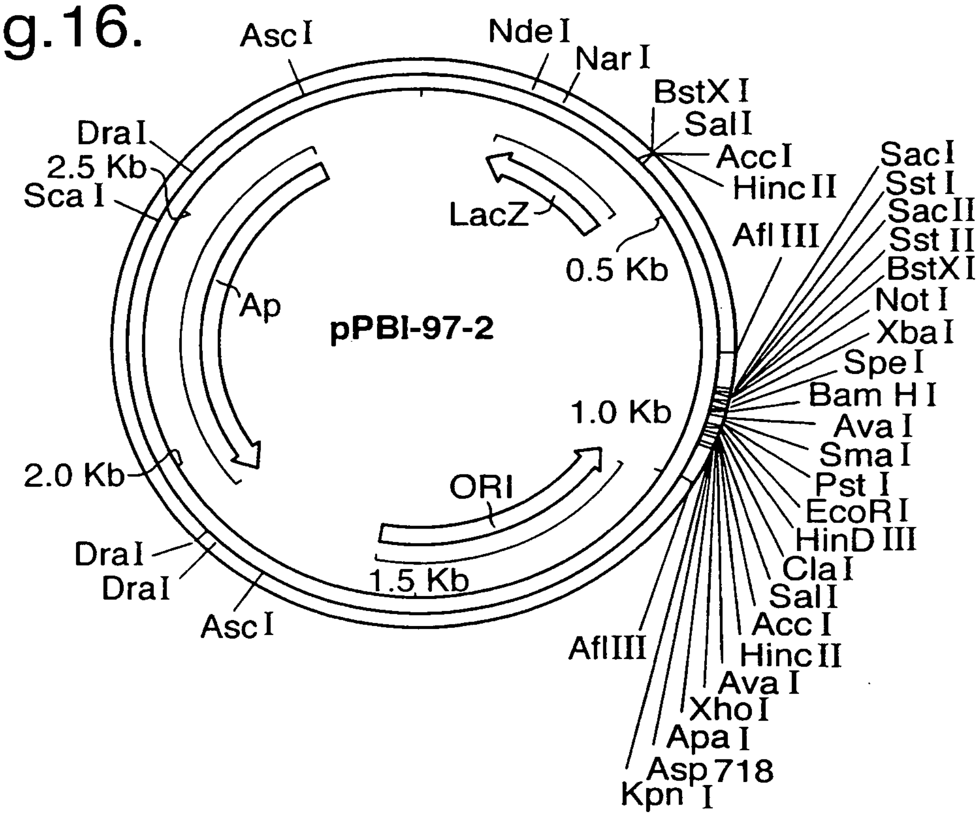

AscI
NdeI
NarI
BstXI
SalI
AccI
HincII
SacI
SstI
SacII
SstII
BstXI
NotI
XbaI
SpeI
Bam HI
AvaI
SmaI
PstI
EcoRI
HinDIII
ClaI
SalI
AccI
HincII
AvaI
XhoI
ApaI
Asp718
Kpn I
AflIII
AflIII
DraI
2.5 Kb
ScaI
LacZ
0.5 Kb
Ap
pPBI-97-2
1.0 Kb
2.0 Kb
ORI
DraI
DraI
1.5 Kb
AscI

Fig.17.

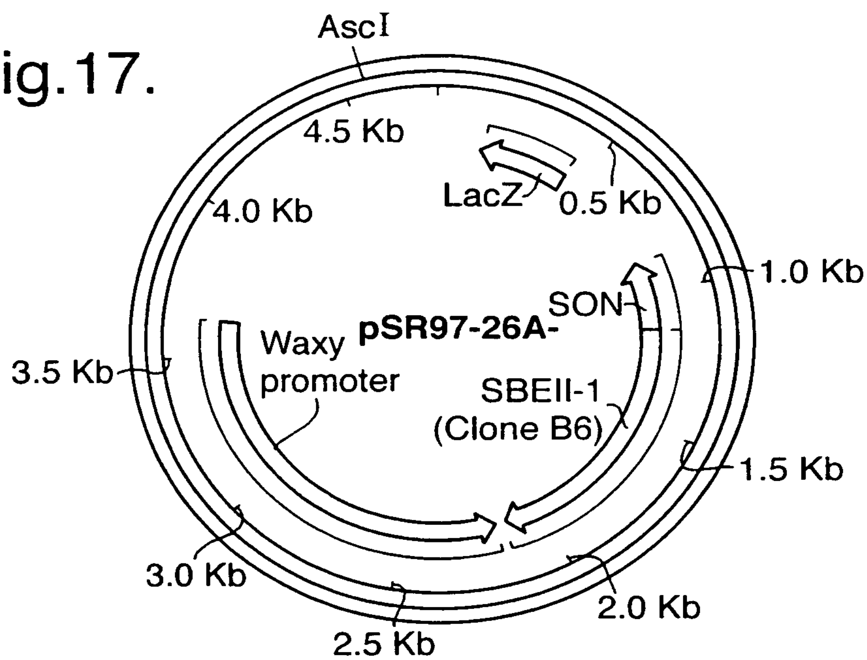

AscI
4.5 Kb
4.0 Kb
LacZ
0.5 Kb
1.0 Kb
SON
pSR97-26A-
Waxy
promoter
3.5 Kb
SBEII-1
(Clone B6)
1.5 Kb
3.0 Kb
2.0 Kb
2.5 Kb

Fig.18.

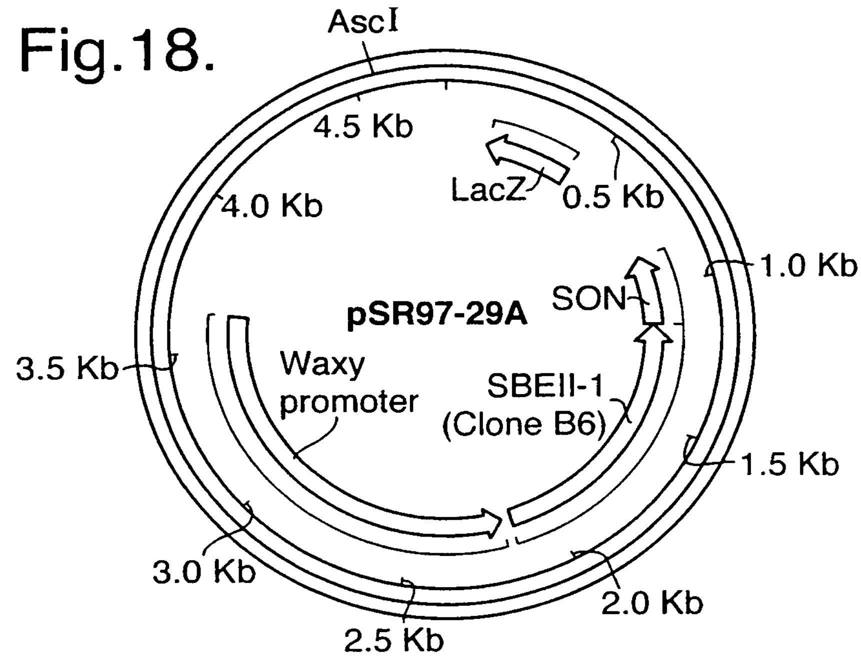

AscI
4.5 Kb
LacZ
0.5 Kb
4.0 Kb
1.0 Kb
SON
pSR97-29A
3.5 Kb
Waxy
promoter
SBEII-1
(Clone B6)
1.5 Kb
3.0 Kb
2.0 Kb
2.5 Kb

Fig.19.

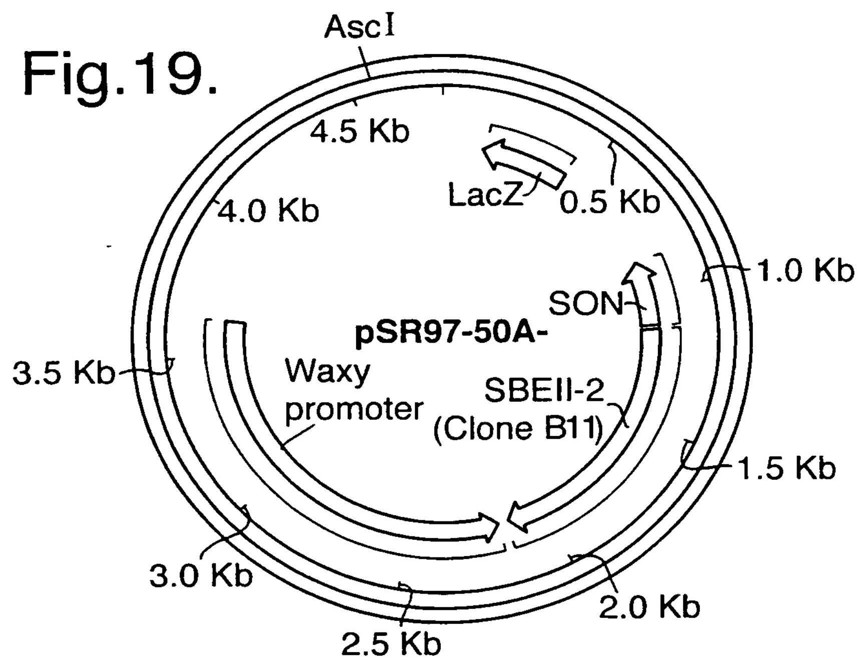

AscI
4.5 Kb
LacZ
0.5 Kb
4.0 Kb
1.0 Kb
SON
pSR97-50A-
3.5 Kb
Waxy
promoter
SBEII-2
(Clone B11)
1.5 Kb
3.0 Kb
2.0 Kb
2.5 Kb

Fig.20.

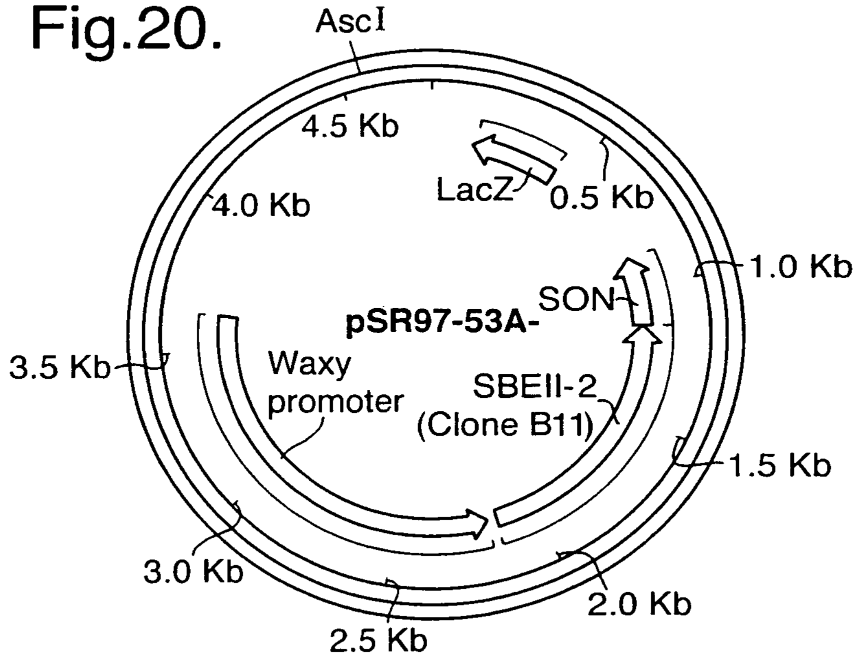

AscI
4.5 Kb
LacZ
0.5 Kb
4.0 Kb
1.0 Kb
SON
pSR97-53A-
Waxy
promoter
SBEII-2
(Clone B11)
3.5 Kb
1.5 Kb
3.0 Kb
2.0 Kb
2.5 Kb

Fig.21.

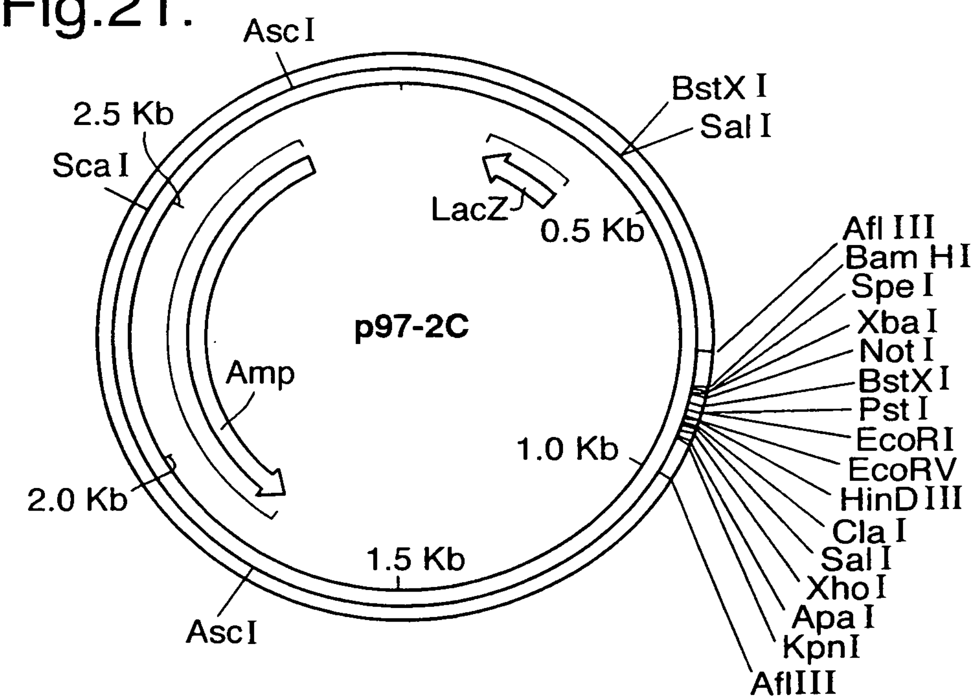

AscI
BstX I
Sal I
2.5 Kb
Sca I
LacZ
0.5 Kb
Afl III
Bam HI
Spe I
Xba I
Not I
BstX I
Pst I
EcoRI
EcoRV
HinD III
Cla I
Sal I
Xho I
Apa I
KpnI
AflIII
p97-2C
Amp
1.0 Kb
2.0 Kb
1.5 Kb
AscI

Fig.22.

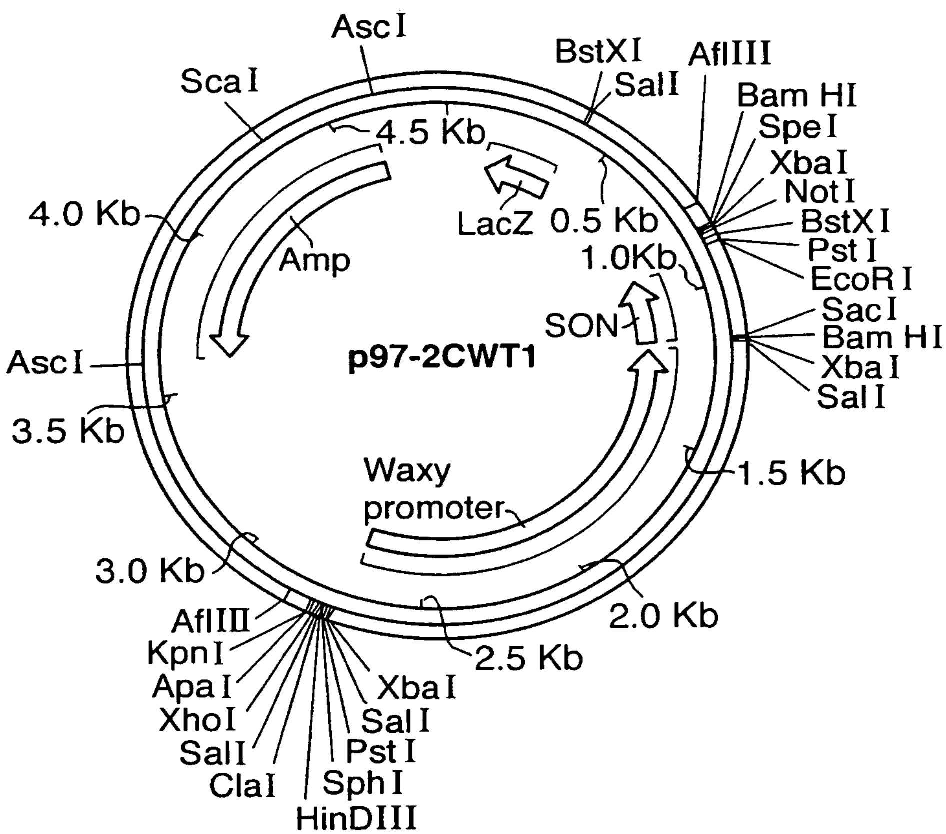

AscI
ScaI
BstXI
SalI
AflIII
Bam HI
SpeI
XbaI
NotI
BstXI
PstI
EcoRI
SacI
Bam HI
XbaI
SalI
4.5 Kb
LacZ
0.5 Kb
1.0Kb
4.0 Kb
Amp
SON
AscI
p97-2CWT1
3.5 Kb
1.5 Kb
Waxy
promoter
3.0 Kb
2.0 Kb
2.5 Kb
AflIII
KpnI
ApaI
XhoI
SalI
ClaI
HinDIII
SphI
PstI
SalI
XbaI

Fig.23.

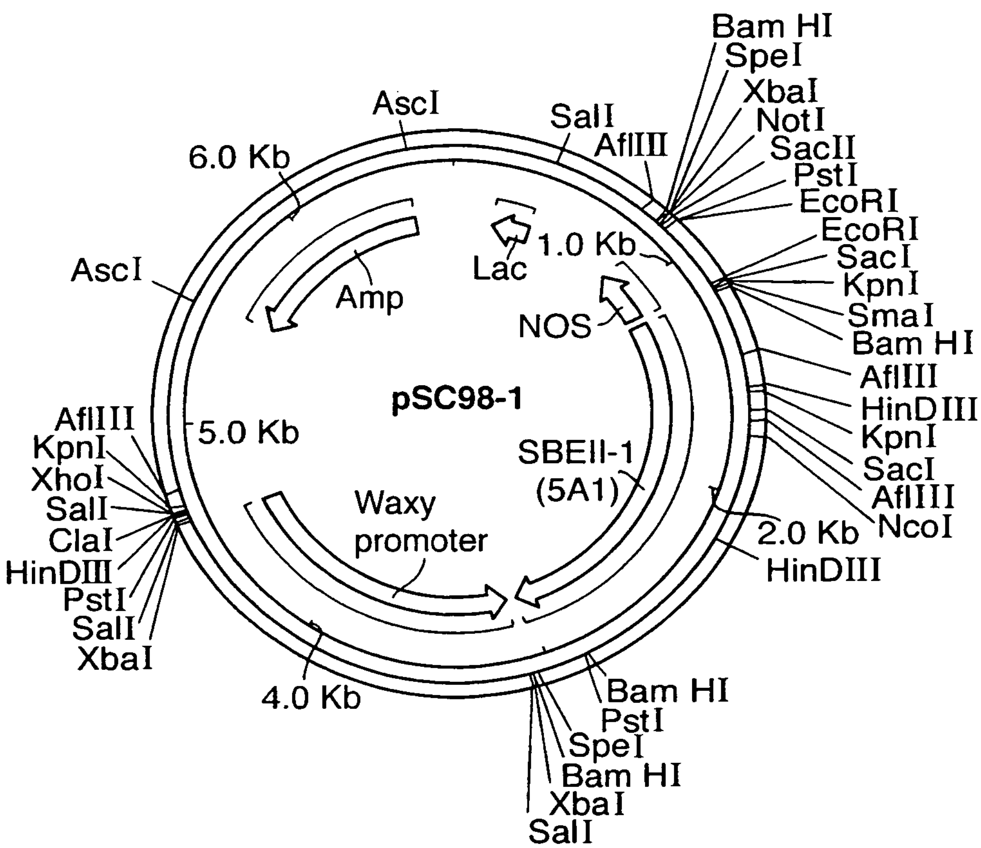

pSC98-1
Amp
Lac
1.0 Kb
NOS
SBEII-1
(5A1)
Waxy
promoter
2.0 Kb
4.0 Kb
5.0 Kb
6.0 Kb
AscI
AscI
SalI
AflIII
Bam HI
SpeI
XbaI
NotI
SacII
PstI
EcoRI
EcoRI
SacI
KpnI
SmaI
Bam HI
AflIII
HinDIII
KpnI
SacI
AflIII
NcoI
HinDIII
Bam HI
PstI
SpeI
Bam HI
XbaI
SalI
AflIII
KpnI
XhoI
SalI
ClaI
HinDIII
PstI
SalI
XbaI

Fig.24.

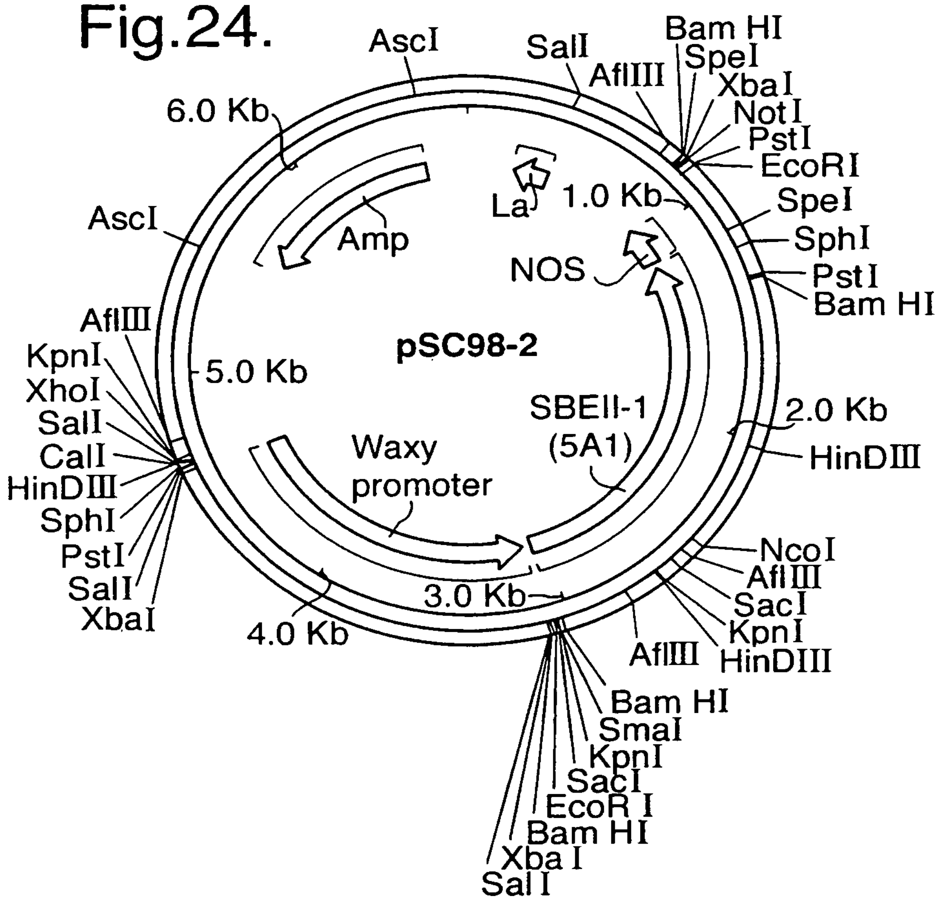

AscI
SalI
AflIII
Bam HI
SpeI
XbaI
NotI
PstI
EcoRI
SpeI
SphI
PstI
Bam HI
6.0 Kb
AscI
Amp
La
1.0 Kb
NOS
pSC98-2
AflIII
KpnI
XhoI
SalI
CalI
HinDIII
SphI
PstI
SalI
XbaI
5.0 Kb
SBEII-1
(5A1)
2.0 Kb
HinDIII
Waxy
promoter
NcoI
AflIII
SacI
KpnI
HinDIII
AflIII
3.0 Kb
4.0 Kb
Bam HI
SmaI
KpnI
SacI
EcoR I
Bam HI
Xba I
Sal I

Fig.25.

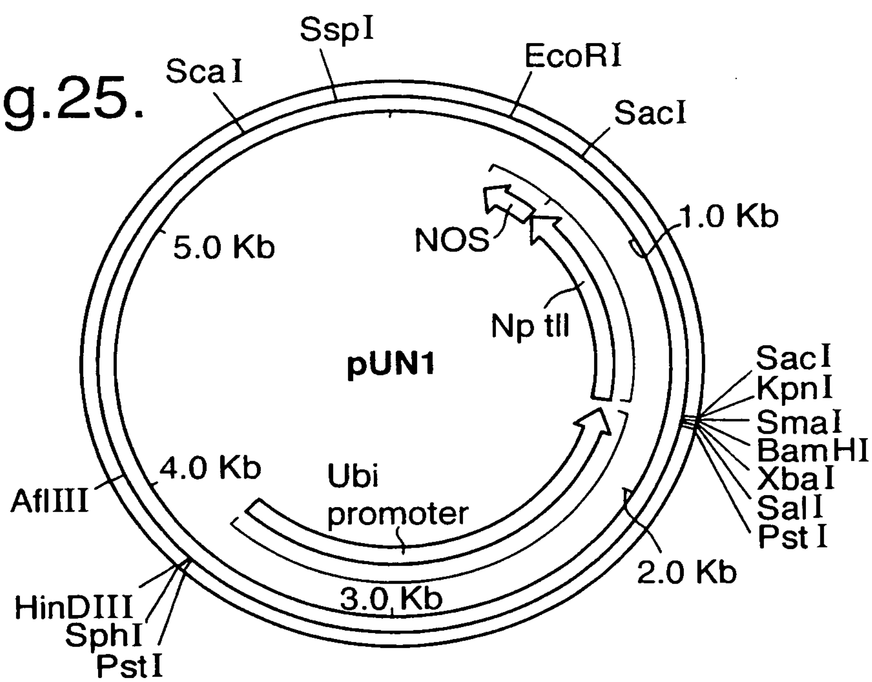

SspI
ScaI
EcoRI
SacI
1.0 Kb
NOS
5.0 Kb
Np tII
pUN1
SacI
KpnI
SmaI
BamHI
XbaI
SalI
PstI
4.0 Kb
AflIII
Ubi
promoter
2.0 Kb
3.0 Kb
HinDIII
SphI
PstI

Fig.26.

```
         10         20         30         40         50         60
GAGCTCCGTT TCGCATGATT GAACAAGATG GATTGCACGC AGGTTCTCCG GCCGCTTGGG 60
TGGAGAGGCT ATTCGGCTAT GACTGGGCAC AACAGACAAT CGGCTGCTCT GATGCCGCCG 120
TGTTCCGGCT GTCAGCGCAG GGGCGCCCGG TTCTTTTTGT CAAGACCGAC CTGTCCGGTG 180
CCCTGAATGA ACTGCAGGAC GAGGCAGCGC GGCTATCGTG GCTGGCCACG ACGGGCGTTC 240
CTTGCGCAGC TGTGCTCGAC GTTGTCACTG AAGCGGGAAG GGACTGGCTG CTATTGGGCG 300
        310        320        330        340        350        360
AAGTGCCGGG GCAGGATCTC CTGTCATCTC ACCTTGCTCC TGCCGAGAAA GTATCCATCA 360
TGGCTGATGC AATGCGGCGG CTGCATACGC TTGATCCGGC TACCTGCCCA TTCGACCACC 420
AAGCGAAACA TCGCATCGAG CGAGCACGTA CTCGGATGGA AGCCGGTCTT GTCGATCAGG 480
ATGATCTGGA CGAAGAGCAT CAGGGGCTCG CGCCAGCCGA ACTGTTCGCC AGGCTCAAGG 540
CGCGCATGCC CGACGGCGAG GATCTCGTCG TGACCCATGG CGATGCCTGC TTGCCGAATA 600
        610        620        630        640        650        660
TCATGGTGGA AAATGGCCGC TTTTCTGGAT TCATCGACTG TGGCCGGCTG GGTGTGGCGG 660
ACCGCTATCA GGACATAGCG TTGGCTACCC GTGATATTGC TGAAGAGCTT GGCGGCGAAT 720
GGGCTGACCG CTTCCTCGTG CTTTACGGTA TCGCCGCTCC CGATTCGCAG CGCATCGCCT 780
TCTATCGCCT TCTTGACGAG TTCTTCTGAG Ctc 813 (SEQ ID No : 35)
```

Fig.27.

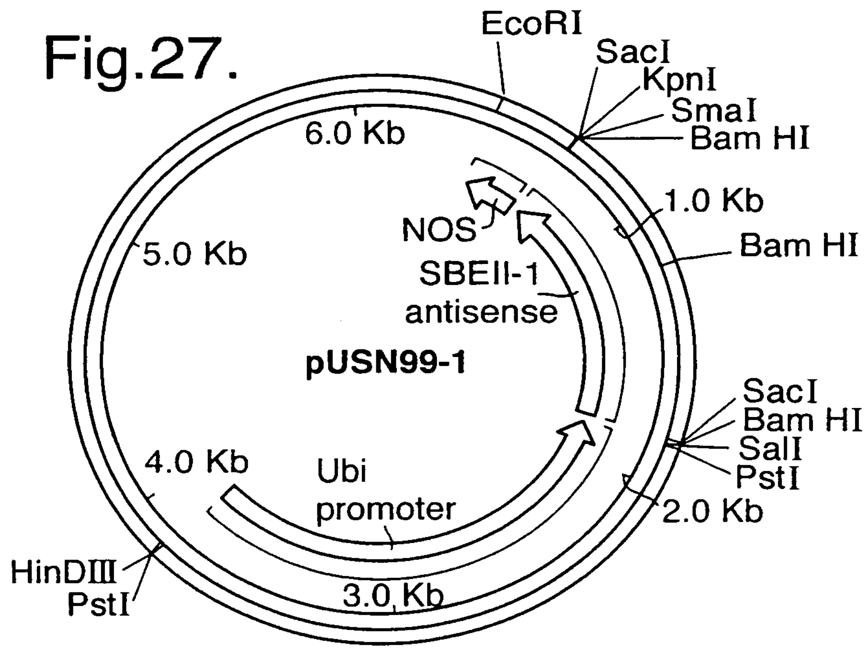

EcoRI
SacI
KpnI
SmaI
Bam HI
1.0 Kb
Bam HI
SacI
Bam HI
SalI
PstI
2.0 Kb
3.0 Kb
4.0 Kb
5.0 Kb
6.0 Kb
HinDIII
PstI
NOS
SBEII-1
antisense
pUSN99-1
Ubi
promoter

Fig.28.

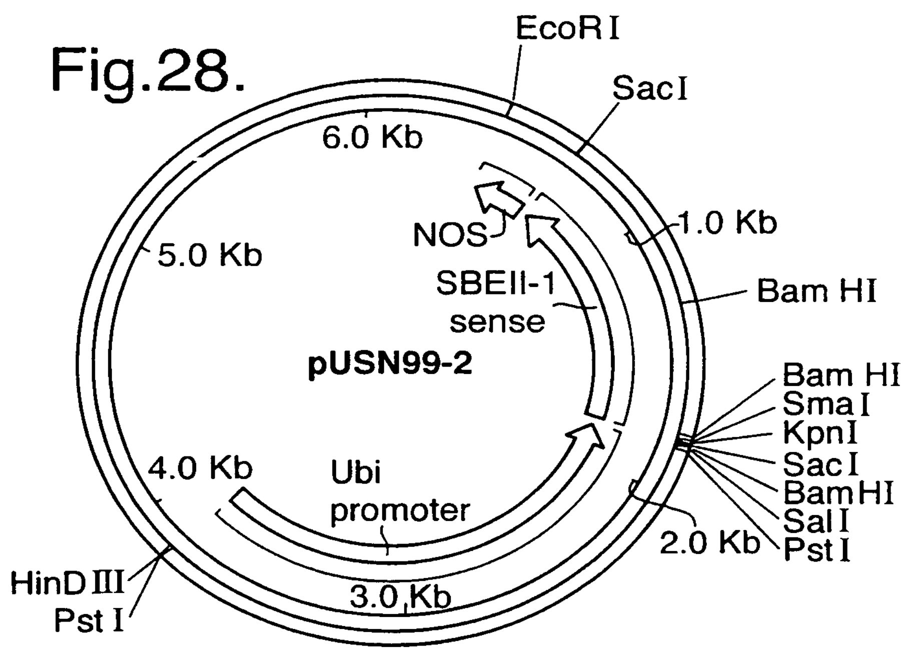

EcoRI
SacI
1.0 Kb
Bam HI
Bam HI
SmaI
KpnI
SacI
BamHI
SalI
PstI
2.0 Kb
3.0 Kb
4.0 Kb
5.0 Kb
6.0 Kb
HinDIII
Pst I
NOS
SBEII-1
sense
pUSN99-2
Ubi
promoter

Fig.29(i).

```
                                    Pst I                                         Xba I
TTAGCTGAATCCGGCGGCATGGCAAGGTAGACTGCAGTGCAGCGTGACCCGGTCGTGCCCCTCTCTAGAGATAATGAGCA
                                                                                   80
AATCGACTTAGGCCGCCGTACCGTTCCATCTGACGTCACGTCGCACTGGGCCAGCACGGGGAGAGATCTCTATTACTCGT

TTGCATGTCTAAGTTATAAAAAAATTACCACATATTTTTTTGTCACACTTGTTTGAAGTGCAGTTTATCTATCTTTATAC
                                                                                   160
AACGTACAGATTCAATATTTTTTTAATGGTGTATAAAAAAACAGTGTGAACAAACTTCACGTCAAATAGATAGAAATATG

                                         Sca I
ATATATTTAAACTTTACTCTACGAATAATATAATCTATAGTACTACAATAATATCAGTGTTTTAGAGAATCATATAAATG
                                                                                   240
TATATAAATTTGAAATGAGATGCTTATTATATTAGATATCATGATGTTATTATAGTCACAAAATCTCTTAGTATATTTAC

AACAGTTAGACATGGTCTAAAGGACAATTGAGTATTTTGACAACAGGACTCTACAGTTTTATCTTTTTAGTGTGCATGTG
                                                                                   320
TTGTCAATCTGTACCAGATTTCCTGTTAACTCATAAAACTGTTGTCCTGAGATGTCAAAATAGAAAAATCACACGTACAC

TTCTCCTTTTTTTTTTTGCAAATAGCTTCACCTATATAATACTTCATCCATTTTATTAGTACATCCATTTAGGGTTTAGG
                                                                                   400
AAGAGGAAAAAAAAAAACGTTTATCGAAGTGGATATATTATGAAGTAGGTAAAATAATCATGTAGGTAAATCCCAAATCC

GTTAATGGTTTTTATAGACTAATTTTTTTAGTACATCTATTTTATTCTATTTTAGCCTCTAAATTAAGAAAACTAAAACT
                                                                                   480
CAATTACCAAAAATATCTGATTAAAAAAATCATGTAGATAAAATAAGATAAAATCGGAGATTTAATTCTTTTGATTTTGA

CTATTTTAGTTTTTTTATTTAATAATTTAGATATAAAATAGAATAAAATAAAGTGACTAAAAATTAAACAAATACCCTTT
                                                                                   560
GATAAAATCAAAAAAATAAATTATTAAATCTATATTTTATCTTATTTTATTTCACTGATTTTTAATTTGTTTATGGGAAA
```

Fig.29(ii).

```
                                                                          Sal I
AAGAAATTAAAAAAACTAAGGAAACATTTTTCTTGTTTCGAGTAGATAATGCCAGCCTGTTAAACGCCGTCGACGCAGTC
-------------------------------------------------------------------------------- 640
TTCTTTAATTTTTTTGATTCCTTTGTAAAAAGAACAAAGCTCATCTATTACGGTCGGACAATTTGCGGCAGCTGCGTCAG

TAACGGACACCAACCAGCGAACCAGCAGCGTCGCGTCGGGCCAAGCGAAGCAGACGGCACGGCATCTCTGTCGCTGCCTC
-------------------------------------------------------------------------------- 720
ATTGCCTGTGGTTGGTCGCTTGGTCGTCGCAGCGCAGCCCGGTTCGCTTCGTCTGCCGTGCCGTAGAGACAGCGACGGAG

Kpn I
GGTACCGGACTTCGTCCGCTGTCGGCATCCAGAAATTGCGTGGCGGAGCGGCAGACGTGAGCCGGCACGGCAGGCGGCCT
-------------------------------------------------------------------------------- 800
CCATGGCCTGAAGCAGGCGACAGCCGTAGGTCTTTAACGCACCGCCTCGCCGTCTGCACTCGGCCGTGCCGTCCGCCGGA

CCTCCTCCTCTCACGGCACCGGCAGCTACGGGGGATTCCTTTCCCACCGCTCCTTCGCTTTCCCTTCCTCGCCCGCCGTA
-------------------------------------------------------------------------------- 880
GGAGGAGGAGAGTGCCGTGGCCGTCGATGCCCCCTAAGGAAAGGGTGGCGAGGAAGCGAAAGGGAAGGAGCGGGCGGCAT

ATAAATAGACACCCCCTCCACACCCTCTTTCCCCAACCTCGTGTTGTTCGGAGCGCACACACACACAACCAGATCTCCCC
-------------------------------------------------------------------------------- 960
TATTTATCTGTGGGGGAGGTGTGGGAGAAAGGGGTTGGAGCACAACAAGCCTCGCGTGTGTGTGTGTTGGTCTAGAGGGG

                                                                       Xba I
CAAATCCACCCGTCGGCACCTCCGCTTCAAGGTACGCCGCTCGTCCTCCCCCCCCCTCTCTACCTTCTCTAGATCGGCGT
-------------------------------------------------------------------------------- 1040
GTTTAGGTGGGCAGCCGTGGAGGCGAAGTTCCATGCGGCGAGCAGGAGGGGGGGGGAGAGATGGAAGAGATCTAGCCGCA

       Nco I
       Sty I        Apa I
TCCGGTCCATGGTTAGGGCCCGGTAGTTCTACTTCTGTTCATGTTTGTGTTAGATCCGTGTTTGTGTTAGATCCGTGCTG
-------------------------------------------------------------------------------- 1120
AGGCCAGGTACCAATCCCGGGCCATCAAGATGAAGACAAGTACAAACACAATCTAGGCACAAACACAATCTAGGCACGAC
```

Fig.29(iii).

```
                                         Afl III
CTAGCGTTCGTACACGGATGCGACCTGTACGTCAGACACGTTCTGATTGCTAACTTGCCAGTGTTTCTCTTTGGGGAATC 1200
GATCGCAAGCATGTGCCTACGCTGGACATGCAGTCTGTGCAAGACTAACGATTGAACGGTCACAAAGAGAAACCCCTTAG

                                       Cla I
CTGGGATGGCTCTAGCCGTTCCGCAGACGGGATCGATTTCATGATTTTTTTTGTTTCGTTGCATAGGGTTTGGTTTGCCC 1280
GACCCTACCGAGATCGGCAAGGCGTCTGCCCTAGCTAAAGTACTAAAAAAAACAAAGCAACGTATCCCAAACCAAACGGG

TTTTCCTTTATTTCAATATATGCCGTGCACTTGTTTGTCGGGTCATCTTTTCATGCTTTTTTTTGTCTTGGTTGTGATGA 1360
AAAAGGAAATAAAGTTATATACGGCACGTGAACAAACAGCCCAGTAGAAAAGTACGAAAAAAAACAGAACCAACACTACT

                  Xba I          EcoR I
TGTGGTCTGGTTGGGCGGTCGTTCTAGATCGGAGTAGAATTCTGTTTCAAACTACCTGGTGGATTTATTAATTTTGGATC 1440
ACACCAGACCAACCCGCCAGCAAGATCTAGCCTCATCTTAAGACAAAGTTTGATGGACCACCTAAATAATTAAAACCTAG

                                                        Cla I           Afl III
TGTATGTGTGTGCCATACATATTCATAGTTACGAATTGAAGATGATGGATGGAAATATCGATCTAGGATAGGTATACATG 1520
ACATACACACACGGTATGTATAAGTATCAATGCTTAACTTCTACTACCTACCTTTATAGCTAGATCCTATCCATATGTAC

                  Nsi I
TTGATGCGGGTTTTACTGATGCATATACAGAGATGCTTTTGTTCGCTTGGTTGTGATGATGTGGTGTGGTTGGGCGGTCG 1600
AACTACGCCCAAAATGACTACGTATATGTCTCTACGAAAACAAGCGAACCAACACTACTACACCACACCAACCCGCCAGC
```

Fig.29(iv).

Xba I

TTCATTCGTTCTAGATCGGAGTAGAATACTGTTTCAAACTACCTGGTGTATTTATTAATTTTGGAACTGTATGTGTGTGT 1680
AAGTAAGCAAGATCTAGCCTCATCTTATGACAAAGTTTGATGGACCACATAAATAATTAAAACCTTGACATACACACACA

Cla I Afl III

CATACATCTTCATAGTTACGAGTTTAAGATGGATGGAAATATCGATCTAGGATAGGTATACATGTTGATGTGGGTTTTAC 1760
GTATGTAGAAGTATCAATGCTCAAATTCTACCTACCTTTATAGCTAGATCCTATCCATATGTACAACTACACCCAAAATG

Nsi I

TGATGCATATACATGATGGCATATGCAGCATCTATTCATATGCTCTAACCTTGAGTACCTATCTATTATAATAAACAAGT 1840
ACTACGTATATGTACTACCGTATACGTCGTAGATAAGTATACGAGATTGGAACTCATGGATAGATAATATTATTTGTTCA

ATGTTTTATAATTATTTTGATCTTGATATACTTGGATGATGGCATATGCAGCAGCTATATGTGGATTTTTTTAGCCCTGC 1920
TACAAAATATTAATAAAACTAGAACTATATGAACCTACTACCGTATACGTCGTCGATATACACCTAAAAAAATCGGGACG

Pst I

CTTCATACGCTATTTATTTGCTTGGTACTGTTTCTTTTGTCGATGCTCACCCTGTTGTTTGGTGTTACTTCTGCAGATGC 2000
GAAGTATGCGATAAATAAACGAACCATGACAAAGAAAACAGCTACGAGTGGGACAACAAACCACAATGAAGACGTCTACG

AGATCTTTGTGAAAACCCTGACTGGCAAGACTATCACC 2038 (SEQ ID No : 52)
TCTAGAAACACTTTTGGGACTGACCGTTCTGATAGTGG

Fig.30.

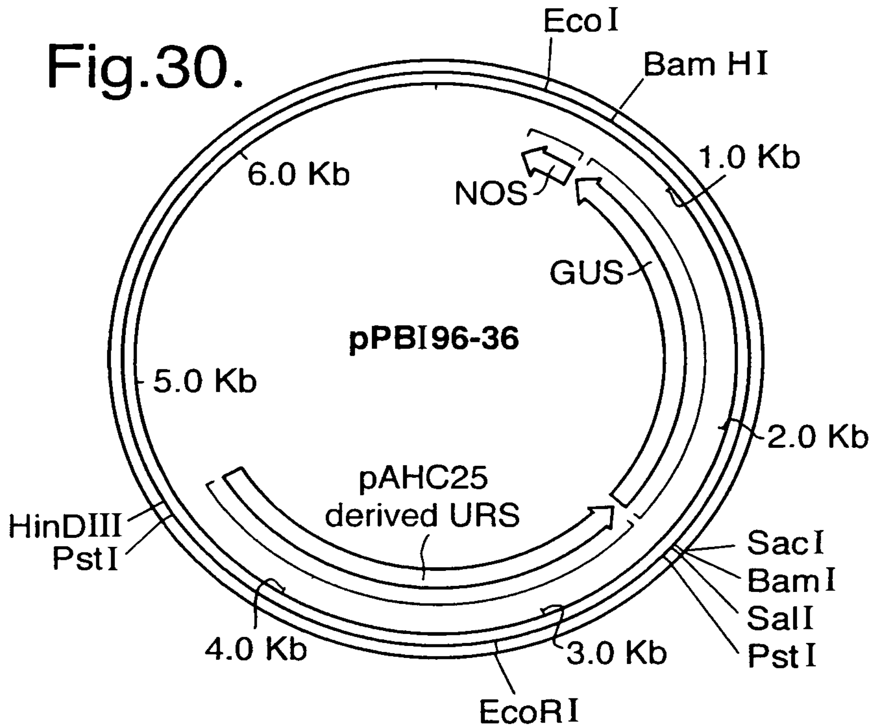

Eco I
Bam HI
1.0 Kb
6.0 Kb
NOS
GUS
pPBI96-36
5.0 Kb
2.0 Kb
pAHC25
derived URS
HinDIII
PstI
SacI
BamI
SalI
PstI
4.0 Kb
3.0 Kb
EcoRI

Fig.31.

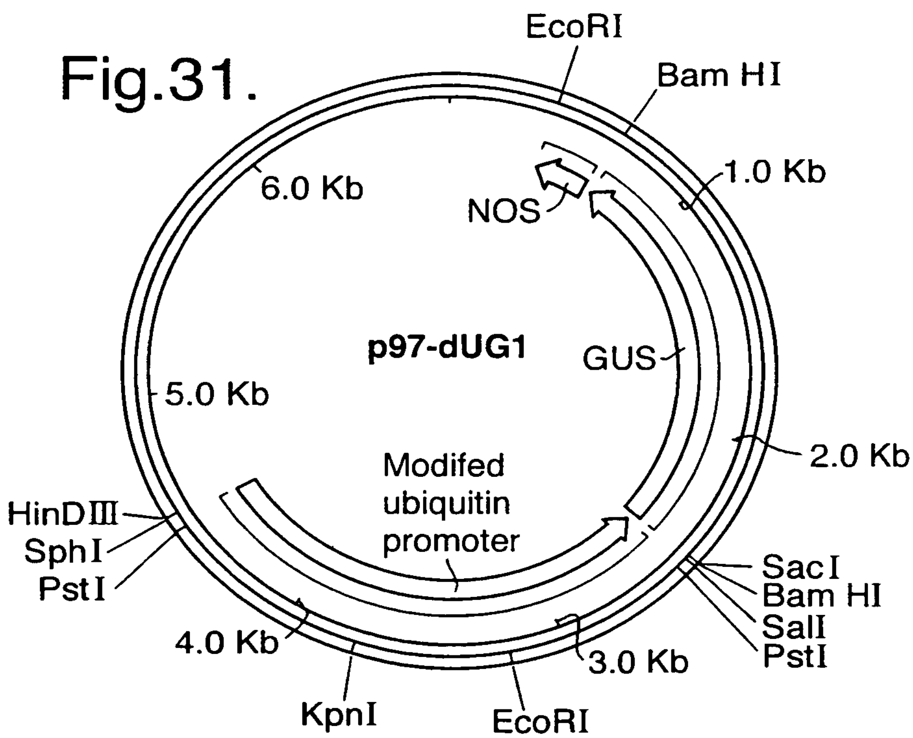

EcoRI
Bam HI
1.0 Kb
6.0 Kb
NOS
p97-dUG1
GUS
5.0 Kb
2.0 Kb
Modifed
ubiquitin
promoter
HinDIII
SphI
PstI
SacI
Bam HI
SalI
PstI
4.0 Kb
3.0 Kb
KpnI
EcoRI

Fig.32.

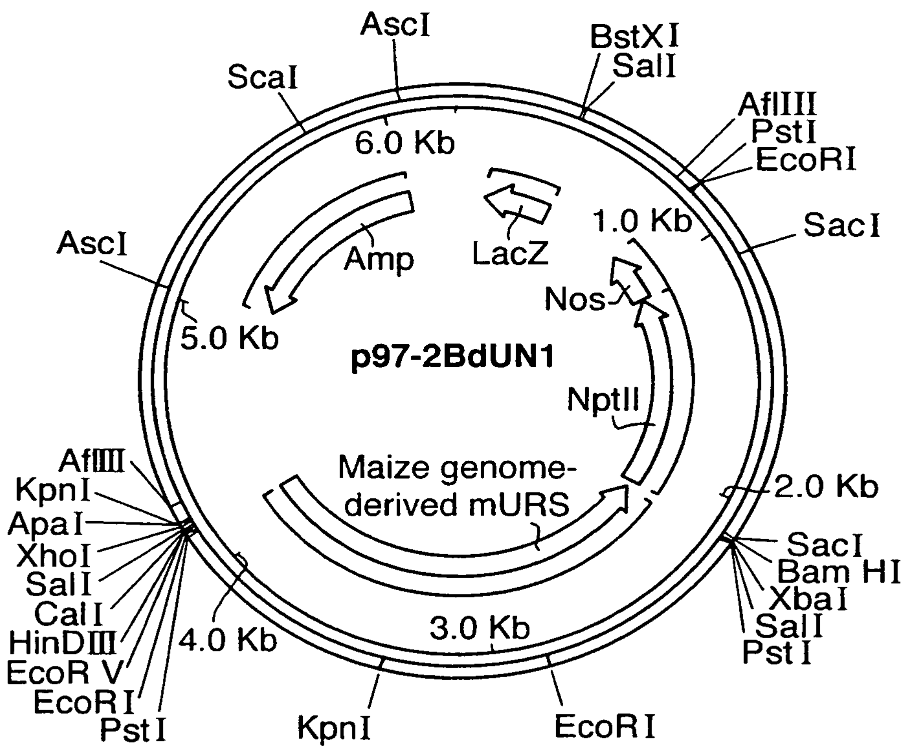

AscI
ScaI
BstXI
SalI
AflIII
PstI
EcoRI
6.0 Kb
1.0 Kb
SacI
AscI
Amp
LacZ
Nos
5.0 Kb
p97-2BdUN1
NptII
AflIII
KpnI
ApaI
XhoI
SalI
CalI
HinDIII
EcoR V
EcoRI
PstI
Maize genome-derived mURS
2.0 Kb
SacI
Bam HI
XbaI
SalI
PstI
4.0 Kb
3.0 Kb
KpnI
EcoRI

Fig.33.

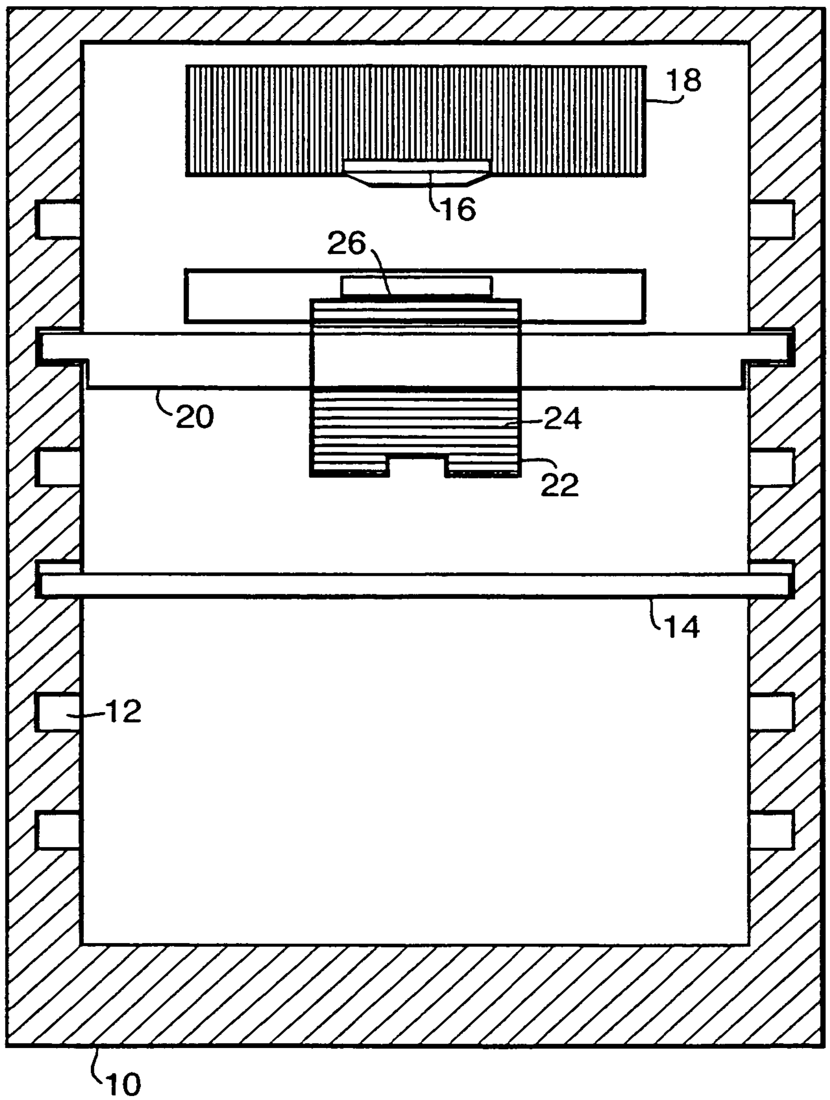
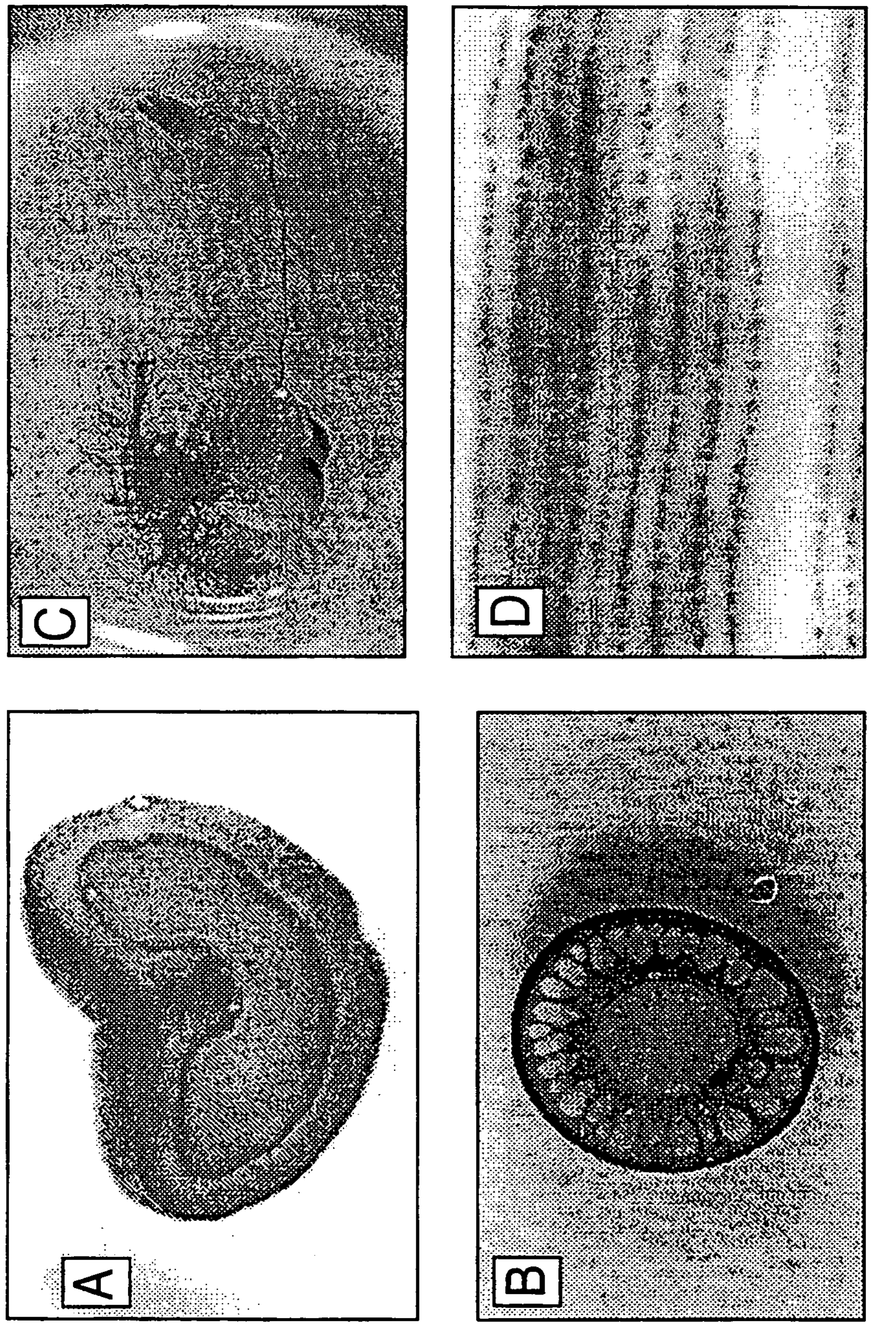


Fig.34.



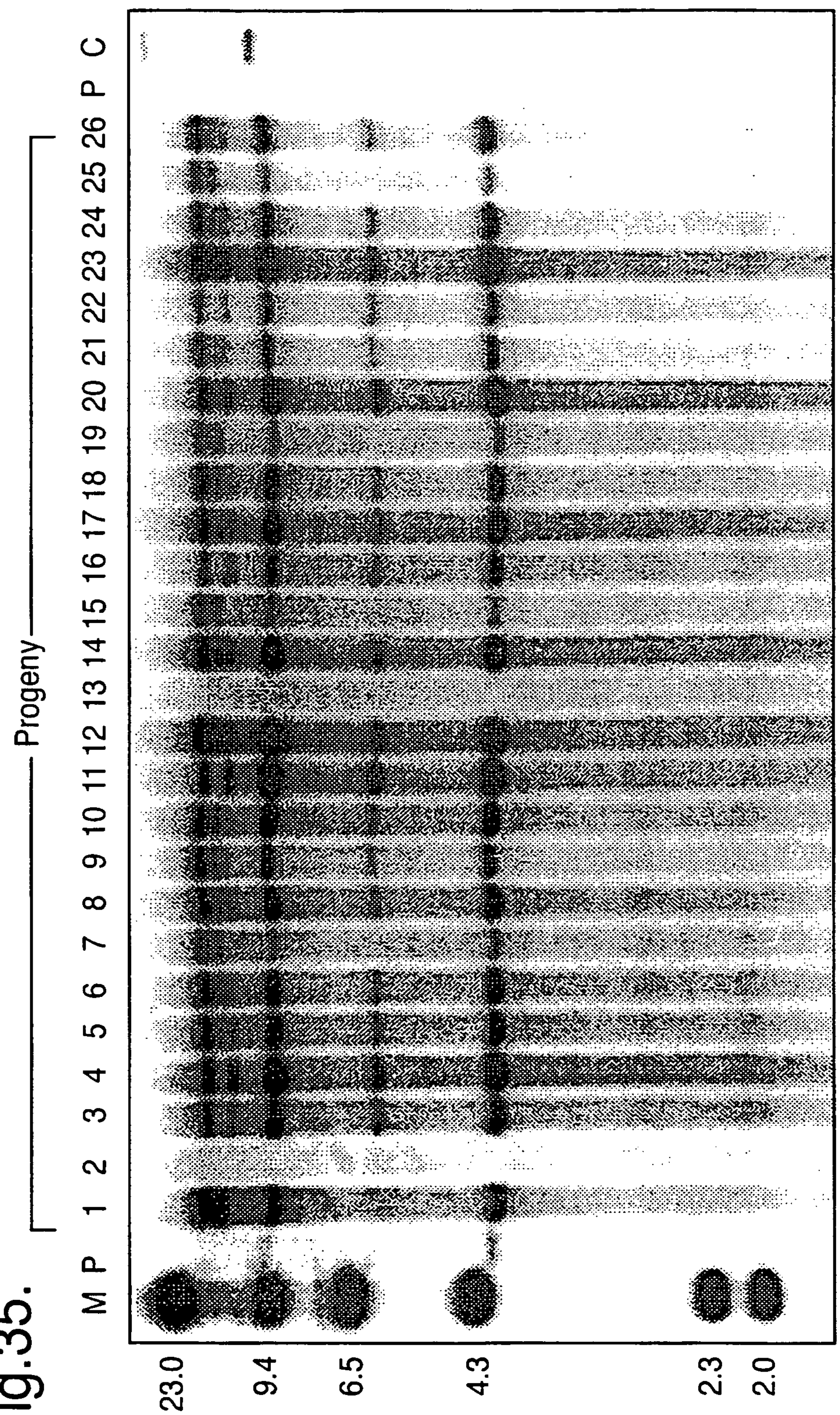

M P 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 P C
Progeny
23.0
9.4
6.5
4.3
2.3
2.0

Fig.35.

Fig.36.

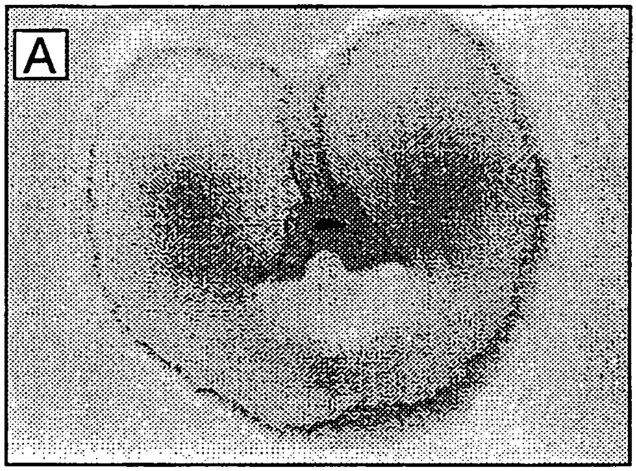
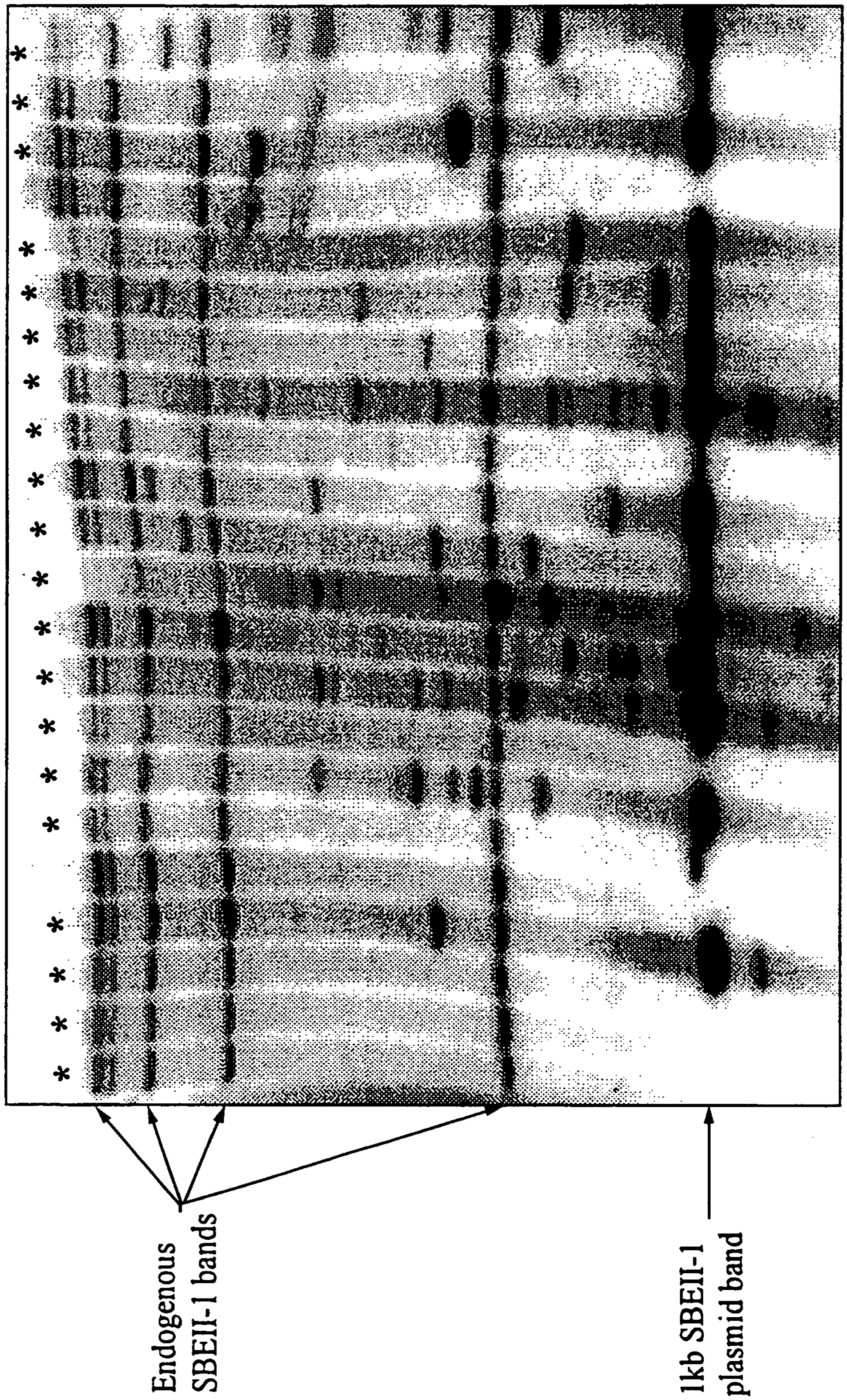


Fig.37.



09/786480
528 Rec'd PCT/PTO 05 MAR 2001

SEQUENCE LISTING

<110> Plant Breeding International Cambridge Limited

<120> Improvements in or relating to plant starch composition

<130> C397.01/U

<140>
<141>

<150> EP 98307337.0
<151> 1998-09-10

<160> 55

<170> PatentIn Ver. 2.1

<210> 1
<211> 2307
<212> DNA
<213> Triticum aestivum

<400> 1
catygacggc cagtgacttc gagctcggta cccggggatc cgatttggtg tgtgggagat 60
gttcttgcca aacaatgcag atggttcgcc accaattcct cacggctcac gggtgaaggt 120
gagaatggat actccatctg ggataaagga ttcaattcct gcttggatca agtactccgt 180
gcagactcca ggagatatac catacaatgg aatatattat gatcctcccg aagaggagaa 240
gtatgtattc aagcatcctc aacctaaacg accaaaatca ttgcggatat atgaaacaca 300
tgttggcatg agtagcccgg aaccaaagat caacacatat gcaaacttca gggatgaggt 360
gcttccaaga attaaaagac ttggatacaa tgcagtgcaa ataatggcaa tccaggagca 420
ctcatactat ggaagctttg ggtaccatgt taccaatttc tttgcaccaa gtagccgttt 480
tgggtcccca gaagatttaa aatctttgat tgatagagct cacgagcttg gcttggttgt 540
cctcatggat gttgttcaca gtcacgcgtc aaataatacc ttggacgggt tgaatggttt 600
tgatggcacg gatacacatt acttccatgg cggttcacgg ggccatcact ggatgtggga 660
ttcccgtgtg tttaactatg ggaataagga agttataagg tttctacttt ccaatgcaag 720
atggtggcta gaggagtata agtttgatgg tttccgattc gatggcgcga cctccatgat 780
gtatacccat catggattac aagtaacctt tacaggaagc taccatgaat attttggctt 840
tgccactgat gtagatgcgg tcgtttactt gatgctgatg aatgatctaa ttcatgggtt 900
ttatcctgaa gccgtaacta tcggtgaaga tgttagtgga atgcctacat ttgcccttcc 960
tgttcaagtt ggtggggttg gttttgacta tcgcttacat atggctgttg ccgacaaatg 1020
gattgaactt ctcaaaggaa acgatgaagc ttgggagatg ggtaatattg tgcacacact 1080
aacaaacaga aggtggccgg aaaagtgtgt tacttatgct gaaagtcacg atcaagcact 1140
ggttggagac aagactattg cattctggtt gatggacaag gatatgtatg atttcatggc 1200
tctgaacgga ccttcgacac ctagtattga tcgtggaata gcactgcata aaatgattag 1260
acttatcaca atgggtttag gaggagaggg ttatcttaac tttatgggaa atgagttcgg 1320
gcatcctgaa tggatagact ttccaagagg cccacaagta cttccaactg gtaagttcat 1380
cccaggaaac aacaacagtt acgacaaatg ccgtcgaaga tttgaccagg gtgatgcaga 1440

```
atttcttagg tatcatggta tgcagcagtt tgatcaggcg atgcagcatc ttgaggaaaa 1500
atatggcttt atgacatcag accaccagta cgtatctcgg aaacatgagg aagataaggt 1560
gatcgtgttt gaaaaagggg acttggtatt tgtgttcaac ttccactgga gtaatagcta 1620
tttcgactac cgggttggct gtttaaagcc tgggaagtac aaggttgtct tagactcaga 1680
cgccggactc tttggtggat ttggtaggat ccatcacact gcagagcact tcacttctga 1740
ctgccaacat gacaacaggc cccattcgtt ctcagtgtac actcctagca gaacctgtgt 1800
tgtctatgct ccaatgaact aaacagcaaa gtgcagcata cgcatgcacg ctgttgttgc 1860
tagcactagc aagaaaaaat cgtatggtca atacaaccag gtgcaaggtt taataagggt 1920
ttgcttcaac gagtcctgga tagacaagac aacatgatga tgtgctctgt gctcccaaat 1980
tcccagggcg ttgtggagaa aaaatgctca tctgtgttat tttatggatc agggangaaa 2040
cctcccccaa anaccccttt ttttttgaa aggnggatag gccccccggtn tctgcatntg 2100
gatgcctcct taaatntttg tagccataaa ccattgctag tgtcctntaa attgacagtt 2160
tagaatagng gttntacttt tgtattttnt ttttgacagt tagactgtat tcctcaaata 2220
atcgacatgt tgtttactcg aagntgagaa ataaaatcag agattgnagn aaaaaaaaaa 2280
aaaaaaaaaa aaaaaaaaaa aaaaaaa                                      2307
```

<210> 2
<211> 758
<212> PRT
<213> Triticum aestivum

<400> 2

```
Ile Asp Gly Gln Leu Arg Ala Arg Tyr Pro Gly Ile Arg Phe Gly Val
1               5                   10                  15

Trp Glu Met Phe Leu Pro Asn Asn Ala Asp Gly Ser Pro Pro Ile Pro
            20                  25                  30

His Gly Ser Arg Val Lys Val Arg Met Asp Thr Pro Ser Gly Ile Lys
        35                  40                  45

Asp Ser Ile Pro Ala Trp Ile Lys Tyr Ser Val Gln Thr Pro Gly Asp
    50                  55                  60

Ile Pro Tyr Asn Gly Ile Tyr Tyr Asp Pro Pro Glu Glu Glu Lys Tyr
65                  70                  75                  80

Val Phe Lys His Pro Gln Pro Lys Arg Pro Lys Ser Leu Arg Ile Tyr
                85                  90                  95

Glu Thr His Val Gly Met Ser Ser Pro Glu Pro Lys Ile Asn Thr Tyr
            100                 105                 110

Ala Asn Phe Arg Asp Glu Val Leu Pro Arg Ile Lys Arg Leu Gly Tyr
        115                 120                 125

Asn Ala Val Gln Ile Met Ala Ile Gln Glu His Ser Tyr Tyr Gly Ser
    130                 135                 140
```

Phe Gly Tyr His Val Thr Asn Phe Phe Ala Pro Ser Ser Arg Phe Gly
145 150 155 160

Ser Pro Glu Asp Leu Lys Ser Leu Ile Asp Arg Ala His Glu Leu Gly
165 170 175

Leu Val Val Leu Met Asp Val Val His Ser His Ala Ser Asn Asn Thr
180 185 190

Leu Asp Gly Leu Asn Gly Phe Asp Gly Thr Asp Thr His Tyr Phe His
195 200 205

Gly Gly Ser Arg Gly His His Trp Met Trp Asp Ser Arg Val Phe Asn
210 215 220

Tyr Gly Asn Lys Glu Val Ile Arg Phe Leu Leu Ser Asn Ala Arg Trp
225 230 235 240

Trp Leu Glu Glu Tyr Lys Phe Asp Gly Phe Arg Phe Asp Gly Ala Thr
245 250 255

Ser Met Met Tyr Thr His His Gly Leu Gln Val Thr Phe Thr Gly Ser
260 265 270

Tyr His Glu Tyr Phe Gly Phe Ala Thr Asp Val Asp Ala Val Val Tyr
275 280 285

Leu Met Leu Met Asn Asp Leu Ile His Gly Phe Tyr Pro Glu Ala Val
290 295 300

Thr Ile Gly Glu Asp Val Ser Gly Met Pro Thr Phe Ala Leu Pro Val
305 310 315 320

Gln Val Gly Gly Val Gly Phe Asp Tyr Arg Leu His Met Ala Val Ala
325 330 335

Asp Lys Trp Ile Glu Leu Leu Lys Gly Asn Asp Glu Ala Trp Glu Met
340 345 350

Gly Asn Ile Val His Thr Leu Thr Asn Arg Arg Trp Pro Glu Lys Cys
355 360 365

Val Thr Tyr Ala Glu Ser His Asp Gln Ala Leu Val Gly Asp Lys Thr
370 375 380

Ile Ala Phe Trp Leu Met Asp Lys Asp Met Tyr Asp Phe Met Ala Leu
385 390 395 400

Asn Gly Pro Ser Thr Pro Ser Ile Asp Arg Gly Ile Ala Leu His Lys
405 410 415

Met Ile Arg Leu Ile Thr Met Gly Leu Gly Gly Glu Gly Tyr Leu Asn
420 425 430

Phe Met Gly Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg
435 440 445

Gly Pro Gln Val Leu Pro Thr Gly Lys Phe Ile Pro Gly Asn Asn Asn
450 455 460

Ser Tyr Asp Lys Cys Arg Arg Arg Phe Asp Gln Gly Asp Ala Glu Phe
465 470 475 480

Leu Arg Tyr His Gly Met Gln Gln Phe Asp Gln Ala Met Gln His Leu
485 490 495

Glu Glu Lys Tyr Gly Phe Met Thr Ser Asp His Gln Tyr Val Ser Arg
500 505 510

Lys His Glu Glu Asp Lys Val Ile Val Phe Glu Lys Gly Asp Leu Val
515 520 525

Phe Val Phe Asn Phe His Trp Ser Asn Ser Tyr Phe Asp Tyr Arg Val
530 535 540

Gly Cys Leu Lys Pro Gly Lys Tyr Lys Val Val Leu Asp Ser Asp Ala
545 550 555 560

Gly Leu Phe Gly Gly Phe Gly Arg Ile His His Thr Ala Glu His Phe
565 570 575

Thr Ser Asp Cys Gln His Asp Asn Arg Pro His Ser Phe Ser Val Tyr
580 585 590

Thr Pro Ser Arg Thr Cys Val Val Tyr Ala Pro Met Asn Thr Ala Lys
595 600 605

Cys Ser Ile Arg Met His Ala Val Val Ala Ser Thr Ser Lys Lys Lys
610 615 620

Ser Tyr Gly Gln Tyr Asn Gln Val Gln Gly Leu Ile Arg Val Cys Phe
625 630 635 640

Asn Glu Ser Trp Ile Asp Lys Thr Thr Cys Ala Leu Cys Ser Gln Ile
645 650 655

```
Pro Arg Ala Leu Trp Arg Lys Asn Ala His Leu Cys Tyr Phe Met Asp
            660                 665                     670

Gln Gly Xaa Asn Leu Pro Gln Xaa Pro Leu Phe Phe Leu Lys Gly Gly
        675                 680                     685

Ala Pro Gly Xaa Cys Xaa Trp Met Pro Pro Xaa Phe Val Ala Ile Asn
    690                 695                     700

His Cys Cys Pro Xaa Asn Gln Phe Arg Ile Xaa Val Xaa Leu Leu Tyr
705                 710                     715                 720

Phe Xaa Phe Asp Ser Thr Val Phe Leu Lys Ser Thr Cys Cys Leu Leu
                725                     730                 735

Glu Xaa Glu Lys Asn Gln Arg Leu Xaa Xaa Lys Lys Lys Lys Lys Lys
            740                 745                     750

Lys Lys Lys Lys Lys Asn
        755
```

<210> 3
<211> 1036
<212> DNA
<213> Triticum aestivum

<400> 3

```
atgtatgatt tcatggctct gaacggacct tcgacgccta atattgatcg tggaatagca 60
ctgcataaaa tgattanact tatcacaatg ggtttaggcg gagagggtta tcttaacttt 120
atgggaaatg agttcgggca tcctgaatgg atagactttc caagaggccc acaagtactt 180
ccaagtggta agttcatccc aggaaacagc aacagttacg acaaatgccg tcgaagattt 240
gacctgggtg atgcagaatt tcttaggtat catggtatgc agcagtttga tcaggcaatg 300
cagcatcttg aggaaaaata tggttttatg acatcagacc accagtacgt atctcggaaa 360
cacgaggaag ataaggtgat cgtgtttgaa aaaggggact tggtatttgt gttcaacttc 420
cactggagta atagctattt cgactaccgg gtcggctgtt taaagcctgg gaagtacaag 480
gtggtcttag actcagacgc tggactcttt ggtggatttg gtaggatcca tcacactgca 540
gagcacttca cttctgactg ccaacatgac aacaggcccc attcgttctc agtgtacact 600
cctagcagaa cctgtgttgt ctatgctcca atgaactaac agcaaggtgc agcatacgcg 660
tgcgcgctgt tgttgctagt agcaagaaaa atcgtacggt caatacagcc aggtgcaagg 720
tttaataagg attttttgct tcaacgagtc ctggatagac aagacaacat gatgttgtgg 780
cgtgtgctcc caatccccag ggcgttgtga agaaaacatg ctcatctgtg ttatgatttt 840
atggatcagc gacgaaactt cccccaaata cccatgcctc cttaaatctt tgtggccgta 900
aaccattgct agtgtcctct aaattgacag tttagcatag aggttttact tttgtatctt 960
ctttttgaca gttagacttt attcctcaaa taatcgacca gtcgtttact cgaaaaaaaa 1020
aaaaaaaaaa aaaaan                                                 1036
```

<210> 4
<211> 1087
<212> DNA
<213> Triticum aestivum

<400> 4
```
atgtatgatt tcatggctct gaacggacct tcgacaccta atattgatcg tggaatagca 60
ctgcataaaa tgattagact tatcacaatg ggtttaggag gagagggtta tcttaacttt 120
atgggaaatg agttcgggca tcctgaatgg atagactttc caagaggccc acaagtactt 180
ccaactggta agttcatccc nngaaacaac aacagttacg acaaatgccg tcgaaaattt 240
gacctgggtg atgcagaatt tcttaggtat catggtatgc agcagtttga tcaggcgatg 300
cagcatcttg aggaaaaata tggctttatg acatcagacc accagtacgt atctcggaaa 360
catgaggaag ataaggtgat cgtgtttgaa aaaggggact tggtatttgt gttcaacttc 420
cactggagta atagctattt cggctaccgg gttggctgtt taaagcctgg gaagtacaag 480
gttgtcttag actcagacgc cggactcttt ggtggatttg gtaggatcca tcacactgca 540
gagcacttca cttctgactg ccaacatgac aacaggcccc attcgttctc agtgtacact 600
cctagcagaa cctgtgttgt ctatgctcca atgaactaaa cagcaaagtg cagcatacgc 660
atgcacgctg ttgttgctag cactagcaag aaaaaatcgt atggtcaata caaccaggtg 720
caaggtttaa taagggtttt tgcttcaacg agtcctggat agacaagaca acatgatgat 780
gtgctctgtg ctcccaattc ccagggcgtt gngnggaaaa catgctcatc tgtgttatc 840
atttatgga tcagngngga aacctccccc aaatacccat gcctccttaa actttgtgg 900
tcctaaacca tggctactat cctctaaatt ggcagtttag catagaggtt ttactttgt 960
aaattttttt tgacagttaa tagactctat tcctcaaata attgacatgt cctttacaag 1020
aagatgagaa ataaaatcag ggattgaaga atcccaaaag ctaaaaaaaa aaaaaaaaaa 1080
aaaaaaa 1087
```

<210> 5
<211> 1120
<212> DNA
<213> Triticum aestivum

<400> 5
```
atgtatgatt tcatggcgct gaacggacct tcgacgccta atattgatcg tggaatagca 60
ctgcataaaa tgattagact tatcacaatg ggtctaggag gagagggtta tcttaacttt 120
atgggaaatg agttcgggca tcctgaatgg atagactttc caagaggccc acaagtactt 180
ccaagtggta agttcatccc aggaaacaac aacagttacg acaaatgccg tcgaagattt 240
gacctgggtg atgcagaatt tcttaggtat catggtatgc agcagtttga tcaggcaatg 300
cagcatcttg aggaaaaata tggttttatg acatcagacc accagtacgt ttctcggaaa 360
catgaggaag ataaggtgat cgtgtttgaa aaaggggact tggtatttgt gttcaacttc 420
cactggagta gtagctattt cgactaccgg gtcggctgtt taaagcctgg gaagtacaag 480
gtggtcttag actcggacgc tggactcttt ggtggatttg gtaggatcca tcacactgca 540
gagcacttca cttctgactg ccaacatgac aacaggcccc attcattctc agtgtacact 600
cctagcagaa cctgtgttgt ctatgctcca atgaactaac agcaaagtgc agcatacgcg 660
tgcgcgctgt tgttgctagt agcaagaaaa atcgtatggt caatacaacc aggtgcaagg 720
tttaataagg atttttgctt caacgagtcc tggatagaca agacaacatg atgttgtgct 780
gtgtgctccc aatccccagg gngttgtgaa gaaaacatgc tcatctgtgt tattttatgg 840
atcagggang aaacctcccc caaanacccc tttttttttt gaaaggngga taggcccccg 900
gtntctgcat ntggatgcct ccttaaatnt ttgtagccat aaaccattgc tagtgtcctn 960
```

taaattgaca gtttagaata gnggttntac ttttgtattt tntttttgac agttagactg 1020
tattcctcaa ataatcgaca tgttgtttac tcgaagntga gaaataaaat cagagattgn 1080
agnaaaaaaa aaaaaaaaaa aaaaaaaaaa aaaaaaaaaa 1120

<210> 6
<211> 979
<212> DNA
<213> Triticum aestivum

<400> 6
atatgtatga tttcatggct ctggataggc cttcaactcc tcgcattgat cgtggcatag 60
cattacataa aatgatcagg cttgtcacca tgggtttagg tggtgaaggc tatcttaact 120
tcatgggaaa tgagtttggg catcctgaat ggatagattt tccaagaggc ccacaaactc 180
ttccaaccgg caaagttctc cctggaaata acaatagtta tgataaatgc cgccatagat 240
ttgatcttgg agatgcagat tttcttagat atcgtggtat gcaagagttc gatcaggcaa 300
tgcagcatct tgaggaaaaa tatgggttta tgacatctga gcaccagtat gtttcacgga 360
aacatgagga agataaggtg atcttcttcg aaagaggaga tttggtattt gttttcaact 420
tccactggag caatagcttt tttgactacc gtgttgggtg ttccaagcct gggaagtaca 480
aggtggcctt ggactccgac gatgcactct ttggtggatt cagcaggctt gatcatgatg 540
tcgactactt cacaaccgaa catccgcatg acaacaggcc gcactctttc tcggtgtaca 600
ctccgagcag aactgcggtc gtgtatgccc ttacagagta agaaccagca gcggcttgtt 660
acaaggcaaa gagagaactc cagagagctc gtggatcgtg agcgaagcga cgggcaacgg 720
cgcgaggctg ctccaagcgc catgactggg aggggatcgt gcntcttccc cagatgccag 780
gaggagcaga tggataggta gcttgttggt gagcgctcga aagaaaatgg acgggcctgg 840
gtgtttgttg tgctgcactg aaccctcctc ctatcttgca cattcccggt tgtttttgta 900
catataacta ataattgccc gtgcgcttca acatgaacat ataaatattc taataggtta 960
aaaaaaaaaa aaaaaaaaa 979

<210> 7
<211> 212
<212> PRT
<213> Triticum aestivum

<400> 7
Met Tyr Asp Phe Met Ala Leu Asn Gly Pro Ser Thr Pro Asn Ile Asp
1 5 10 15

Arg Gly Ile Ala Leu His Lys Met Ile Arg Leu Ile Thr Met Gly Leu
20 25 30

Gly Gly Glu Gly Tyr Leu Asn Phe Met Gly Asn Glu Phe Gly His Pro
35 40 45

Glu Trp Ile Asp Phe Pro Arg Gly Pro Gln Val Leu Pro Ser Gly Lys
50 55 60

Phe Ile Pro Gly Asn Ser Asn Ser Tyr Asp Lys Cys Arg Arg Arg Phe
65 70 75 80

Asp Leu Gly Asp Ala Glu Phe Leu Arg Tyr His Gly Met Gln Gln Phe
85 90 95

Asp Gln Ala Met Gln His Leu Glu Glu Lys Tyr Gly Phe Met Thr Ser
100 105 110

Asp His Gln Tyr Val Ser Arg Lys His Glu Glu Asp Lys Val Ile Val
115 120 125

Phe Glu Lys Gly Asp Leu Val Phe Val Phe Asn Phe His Trp Ser Asn
130 135 140

Ser Tyr Phe Asp Tyr Arg Val Gly Cys Leu Lys Pro Gly Lys Tyr Lys
145 150 155 160

Val Val Leu Asp Ser Asp Ala Gly Leu Phe Gly Gly Phe Gly Arg Ile
165 170 175

His His Thr Ala Glu His Phe Thr Ser Asp Cys Gln His Asp Asn Arg
180 185 190

Pro His Ser Phe Ser Val Tyr Thr Pro Ser Arg Thr Cys Val Val Tyr
195 200 205

Ala Pro Met Asn
210

<210> 8
<211> 378
<212> DNA
<213> Triticum aestivum

<400> 8
actaacagca aggtgcagca tacgcgtgcg cgctgttgtt gctagtagca agaaaaatcg 60
tacggtcaat acagccaggt gcaaggttta ataaggattt tttgcttcaa cgagtcctgg 120
atagacaaga caacatgatg ttgtggcgtg tgctcccaat ccccagggcg ttgtgaagaa 180
aacatgctca tctgtgttat gattttatgg atcagcgacg aaacttcccc caaataccca 240
tgcctcctta aatctttgtg gccgtaaacc attgctagtg tcctctaaat tgacagttta 300
gcatagaggt tttacttttg tatcttcttt ttgacagtta gactttattc ctcaaataat 360
cgaccagtcg tttactcg 378

<210> 9
<211> 449
<212> DNA
<213> Triticum aestivum

<400> 9
aactaacagc aaagtgcagc atacgcgtgc gcgctgttgt tgctagtagc aagaaaaatc 60
gtatggtcaa tacaaccagg tgcaaggttt aataaggatt tttgcttcaa cgagtcctgg 120
atagacaaga caacatgatg ttgtgctgtg tgctcccaat ccccagggng ttgtgaagaa 180
aacatgctca tctgtgttat tttatggatc agggangaaa cctcccccaa anaccccttt 240
ttttttgaa aggnggatag gcccccggtn tctgcatntg gatgcctcct taaatntttg 300
tagccataaa ccattgctag tgtcctntaa attgacagtt tagaatagng gttntacttt 360
tgtattttnt ttttgacagt tagactgtat tcctcaaata atcgacatgt tgtttactcg 420
aagntgagaa ataaaatcag agattgnag 449

<210> 10
<211> 428
<212> DNA
<213> Triticum aestivum

<400> 10
actaaacagc aaagtgcagc atacgcatgc acgctgttgt tgctagcact agcaagaaaa 60
aatcgtatgg tcaatacaac caggtgcaag gtttaataag ggtttttgct tcaacgagtc 120
ctggatagac aagacaacat gatgatgtgc tctgtgctcc caaattccca gggcgttgng 180
nggaaaacat gctcatctgt gttatcattt tatggatcag ngnggaaacc tcccccaaat 240
acccatgcct ccttaaactt ttgtggtcct aaaccatggc tactatcctc taaattggca 300
gtttagcata gaggttttac ttttgtaaat ttttttgac agttaataga ctctattcct 360
caaataattg acatgtcctt tacaagaaga tgagaaataa aatcagggat tgaagaatcc 420
caaaagct 428

<210> 11
<211> 592
<212> PRT
<213> Triticum aestivum

<400> 11
Phe Gly Val Trp Glu Met Phe Leu Pro Asn Asn Ala Asp Gly Ser Pro
1 5 10 15

Pro Ile Pro His Gly Ser Arg Val Lys Val Arg Met Asp Thr Pro Ser
20 25 30

Gly Ile Lys Asp Ser Ile Pro Ala Trp Ile Lys Tyr Ser Val Gln Thr
35 40 45

Pro Gly Asp Ile Pro Tyr Asn Gly Ile Tyr Tyr Asp Pro Pro Glu Glu
50 55 60

Glu Lys Tyr Val Phe Lys His Pro Gln Pro Lys Arg Pro Lys Ser Leu
65 70 75 80

Arg Ile Tyr Glu Thr His Val Gly Met Ser Ser Pro Glu Pro Lys Ile
85 90 95

```
Asn Thr Tyr Ala Asn Phe Arg Asp Glu Val Leu Pro Arg Ile Lys Arg
            100                 105                 110

Leu Gly Tyr Asn Ala Val Gln Ile Met Ala Ile Gln Glu His Ser Tyr
        115                 120                 125

Tyr Gly Ser Phe Gly Tyr His Val Thr Asn Phe Phe Ala Pro Ser Ser
    130                 135                 140

Arg Phe Gly Ser Pro Glu Asp Leu Lys Ser Leu Ile Asp Arg Ala His
145                 150                 155                 160

Glu Leu Gly Leu Val Val Leu Met Asp Val Val His Ser His Ala Ser
                165                 170                 175

Asn Asn Thr Leu Asp Gly Leu Asn Gly Phe Asp Gly Thr Asp Thr His
            180                 185                 190

Tyr Phe His Gly Gly Ser Arg Gly His His Trp Met Trp Asp Ser Arg
        195                 200                 205

Val Phe Asn Tyr Gly Asn Lys Glu Val Ile Arg Phe Leu Leu Ser Asn
    210                 215                 220

Ala Arg Trp Trp Leu Glu Glu Tyr Lys Phe Asp Gly Phe Arg Phe Asp
225                 230                 235                 240

Gly Ala Thr Ser Met Met Tyr Thr His His Gly Leu Gln Val Thr Phe
                245                 250                 255

Thr Gly Ser Tyr His Glu Tyr Phe Gly Phe Ala Thr Asp Val Asp Ala
            260                 265                 270

Val Val Tyr Leu Met Leu Met Asn Asp Leu Ile His Gly Phe Tyr Pro
        275                 280                 285

Glu Ala Val Thr Ile Gly Glu Asp Val Ser Gly Met Pro Thr Phe Ala
    290                 295                 300

Leu Pro Val Gln Val Gly Gly Val Gly Phe Asp Tyr Arg Leu His Met
305                 310                 315                 320

Ala Val Ala Asp Lys Trp Ile Glu Leu Leu Lys Gly Asn Asp Glu Ala
                325                 330                 335

Trp Glu Met Gly Asn Ile Val His Thr Leu Thr Asn Arg Arg Trp Pro
            340                 345                 350
```

Glu Lys Cys Val Thr Tyr Ala Glu Ser His Asp Gln Ala Leu Val Gly
355 360 365

Asp Lys Thr Ile Ala Phe Trp Leu Met Asp Lys Asp Met Tyr Asp Phe
370 375 380

Met Ala Leu Asn Gly Pro Ser Thr Pro Ser Ile Asp Arg Gly Ile Ala
385 390 395 400

Leu His Lys Met Ile Arg Leu Ile Thr Met Gly Leu Gly Gly Glu Gly
405 410 415

Tyr Leu Asn Phe Met Gly Asn Glu Phe Gly His Pro Glu Trp Ile Asp
420 425 430

Phe Pro Arg Gly Pro Gln Val Leu Pro Thr Gly Lys Phe Ile Pro Gly
435 440 445

Asn Asn Asn Ser Tyr Asp Lys Cys Arg Arg Arg Phe Asp Gln Gly Asp
450 455 460

Ala Glu Phe Leu Arg Tyr His Gly Met Gln Gln Phe Asp Gln Ala Met
465 470 475 480

Gln His Leu Glu Glu Lys Tyr Gly Phe Met Thr Ser Asp His Gln Tyr
485 490 495

Val Ser Arg Lys His Glu Glu Asp Lys Val Ile Val Phe Glu Lys Gly
500 505 510

Asp Leu Val Phe Val Phe Asn Phe His Trp Ser Asn Ser Tyr Phe Asp
515 520 525

Tyr Arg Val Gly Cys Leu Lys Pro Gly Lys Tyr Lys Val Val Leu Asp
530 535 540

Ser Asp Ala Gly Leu Phe Gly Gly Phe Gly Arg Ile His His Thr Ala
545 550 555 560

Glu His Phe Thr Ser Asp Cys Gln His Asp Asn Arg Pro His Ser Phe
565 570 575

Ser Val Tyr Thr Pro Ser Arg Thr Cys Val Val Tyr Ala Pro Met Asn
580 585 590

<210> 12
<211> 771
<212> PRT
<213> Triticum aestivum

<400> 12
Ser Arg Ala Ala Ser Pro Gly Lys Val Leu Val Pro Asp Gly Glu Ser
1 5 10 15

Asp Asp Leu Ala Ser Pro Ala Gln Pro Glu Glu Leu Gln Ile Pro Glu
20 25 30

Asp Ile Glu Glu Gln Thr Ala Glu Val Asn Met Thr Gly Gly Thr Ala
35 40 45

Glu Lys Leu Glu Ser Ser Glu Pro Thr Gln Gly Ile Val Glu Thr Ile
50 55 60

Thr Asp Gly Val Thr Lys Gly Val Lys Glu Leu Val Val Gly Glu Lys
65 70 75 80

Pro Arg Val Val Pro Lys Pro Gly Asp Gly Gln Lys Ile Tyr Glu Ile
85 90 95

Asp Pro Thr Leu Lys Asp Phe Arg Ser His Leu Asp Tyr Arg Tyr Ser
100 105 110

Glu Tyr Arg Arg Ile Arg Ala Ala Ile Asp Gln His Glu Gly Gly Leu
115 120 125

Glu Ala Phe Ser Arg Gly Tyr Glu Lys Leu Gly Phe Thr Arg Ser Ala
130 135 140

Glu Gly Ile Thr Tyr Arg Glu Trp Ala Pro Gly Ala His Ser Ala Ala
145 150 155 160

Leu Val Gly Asp Phe Asn Asn Trp Asn Pro Asn Ala Asp Thr Met Thr
165 170 175

Arg Asp Asp Tyr Gly Val Trp Glu Ile Phe Leu Pro Asn Asn Ala Asp
180 185 190

Gly Ser Pro Ala Ile Pro His Gly Ser Arg Val Lys Ile Arg Met Asp
195 200 205

Thr Pro Ser Gly Val Lys Asp Ser Ile Ser Ala Trp Ile Lys Phe Ser

210 215 220

Val Gln Ala Pro Gly Glu Ile Pro Phe Asn Gly Ile Tyr Tyr Asp Pro
225 230 235 240

Pro Glu Glu Glu Lys Tyr Val Phe Gln His Pro Gln Pro Lys Arg Pro
245 250 255

Glu Ser Leu Arg Ile Tyr Glu Ser His Ile Gly Met Ser Ser Pro Glu
260 265 270

Pro Lys Ile Asn Ser Tyr Ala Asn Phe Arg Asp Glu Val Leu Pro Arg
275 280 285

Ile Lys Arg Leu Gly Tyr Asn Ala Val Gln Ile Met Ala Ile Gln Glu
290 295 300

His Ser Tyr Tyr Ala Ser Phe Gly Tyr His Val Thr Asn Phe Phe Ala
305 310 315 320

Pro Ser Ser Arg Phe Gly Thr Pro Glu Asp Leu Lys Ser Leu Ile Asp
325 330 335

Arg Ala His Glu Leu Gly Leu Ile Val Leu Met Asp Ile Val His Ser
340 345 350

His Ser Ser Asn Asn Thr Leu Asp Gly Leu Asn Gly Phe Asp Gly Thr
355 360 365

Asp Thr His Tyr Phe His Gly Gly Pro Arg Gly His His Trp Met Trp
370 375 380

Asp Ser Arg Leu Phe Asn Tyr Gly Ser Trp Glu Val Leu Arg Phe Leu
385 390 395 400

Leu Ser Asn Ala Arg Trp Trp Leu Glu Glu Tyr Lys Phe Asp Gly Phe
405 410 415

Arg Phe Asp Gly Val Thr Ser Met Met Tyr Thr His His Gly Leu Gln
420 425 430

Met Thr Phe Thr Gly Asn Tyr Gly Glu Tyr Phe Gly Phe Ala Thr Asp
435 440 445

Val Asp Ala Val Val Tyr Leu Met Leu Val Asn Asp Leu Ile His Gly
450 455 460

Leu His Pro Asp Ala Val Ser Ile Gly Glu Asp Val Ser Gly Met Pro

465 470 475 480

Thr Phe Cys Ile Pro Val Pro Asp Gly Gly Val Gly Leu Asp Tyr Arg
485 490 495

Leu His Met Ala Val Ala Asp Lys Trp Ile Glu Leu Leu Lys Gln Ser
500 505 510

Asp Glu Ser Trp Lys Met Gly Asp Ile Val His Thr Leu Thr Asn Arg
515 520 525

Arg Trp Leu Glu Lys Cys Val Thr Tyr Ala Glu Ser His Asp Gln Ala
530 535 540

Leu Val Gly Asp Lys Thr Ile Ala Phe Trp Leu Met Asp Lys Asp Met
545 550 555 560

Tyr Asp Phe Met Ala Leu Asp Arg Pro Ser Thr Pro Arg Ile Asp Arg
565 570 575

Gly Ile Ala Leu His Lys Met Ile Arg Leu Val Thr Met Gly Leu Gly
580 585 590

Gly Glu Gly Tyr Leu Asn Phe Met Gly Asn Glu Phe Gly His Pro Glu
595 600 605

Trp Ile Asp Phe Pro Arg Gly Pro Gln Thr Leu Pro Thr Gly Lys Val
610 615 620

Leu Pro Gly Asn Asn Asn Ser Tyr Asp Lys Cys Arg Arg Arg Phe Asp
625 630 635 640

Leu Gly Asp Ala Asp Phe Leu Arg Tyr His Gly Met Gln Glu Phe Asp
645 650 655

Gln Ala Met Gln His Leu Glu Glu Lys Tyr Gly Phe Met Thr Ser Glu
660 665 670

His Gln Tyr Val Ser Arg Lys His Glu Glu Asp Lys Val Ile Ile Phe
675 680 685

Glu Arg Gly Asp Leu Val Phe Val Phe Asn Phe His Trp Ser Asn Ser
690 695 700

Phe Phe Asp Tyr Arg Val Gly Cys Ser Arg Pro Gly Lys Tyr Lys Val
705 710 715 720

Ala Leu Asp Ser Asp Asp Ala Leu Phe Gly Gly Phe Ser Arg Leu Asp

725 730 735

His Asp Val Asp Tyr Phe Thr Thr Glu His Pro His Asp Asn Arg Pro
740 745 750

Arg Ser Phe Ser Val Tyr Thr Pro Ser Arg Thr Ala Val Val Tyr Ala
755 760 765

Leu Thr Glu
770

<210> 13
<211> 797
<212> PRT
<213> Zea mays

<400> 13
Ser Cys Ala Gly Ala Pro Gly Lys Val Leu Val Pro Gly Gly Gly Ser
1 5 10 15

Asp Asp Leu Leu Ser Ser Ala Glu Pro Val Val Asp Thr Gln Pro Glu
20 25 30

Glu Leu Gln Ile Pro Glu Ala Glu Leu Thr Val Glu Lys Thr Ser Ser
35 40 45

Ser Pro Thr Gln Thr Thr Ser Ala Val Ala Glu Ala Ser Ser Gly Val
50 55 60

Glu Ala Glu Glu Arg Pro Glu Leu Ser Ser Glu Val Ile Gly Val Gly
65 70 75 80

Gly Thr Gly Gly Thr Lys Ile Asp Gly Ala Gly Ile Lys Ala Lys Ala
85 90 95

Pro Leu Val Glu Glu Lys Pro Arg Val Ile Pro Pro Pro Gly Asp Gly
100 105 110

Gln Arg Ile Tyr Glu Ile Asp Pro Met Leu Glu Gly Phe Arg Gly His
115 120 125

Leu Asp Tyr Arg Tyr Ser Glu Tyr Lys Arg Leu Arg Ala Ala Ile Asp
130 135 140

Gln His Glu Gly Gly Leu Asp Ala Phe Ser Arg Gly Tyr Glu Lys Leu
145 150 155 160

Gly Phe Thr Arg Ser Ala Glu Gly Ile Thr Tyr Arg Glu Trp Ala Pro
165 170 175

Gly Ala Tyr Ser Ala Ala Leu Val Gly Asp Phe Asn Asn Trp Asn Pro
180 185 190

Asn Ala Asp Ala Met Ala Arg Asn Glu Tyr Gly Val Trp Glu Ile Phe
195 200 205

Leu Pro Asn Asn Ala Asp Gly Ser Pro Ala Ile Pro His Gly Ser Arg
210 215 220

Val Lys Ile Arg Met Asp Thr Pro Ser Gly Val Lys Asp Ser Ile Pro
225 230 235 240

Ala Trp Ile Lys Phe Ser Val Gln Ala Pro Gly Glu Ile Pro Tyr Asn
245 250 255

Gly Ile Tyr Tyr Asp Pro Pro Glu Glu Glu Lys Tyr Val Phe Lys His
260 265 270

Pro Gln Pro Lys Arg Pro Lys Ser Leu Arg Ile Tyr Glu Ser His Val
275 280 285

Gly Met Ser Ser Pro Glu Pro Lys Ile Asn Thr Tyr Ala Asn Phe Arg
290 295 300

Asp Glu Val Leu Pro Arg Ile Lys Lys Leu Gly Tyr Asn Ala Val Gln
305 310 315 320

Ile Met Ala Ile Gln Glu His Ser Tyr Tyr Ala Ser Phe Gly Tyr His
325 330 335

Val Thr Asn Phe Phe Ala Pro Ser Ser Arg Phe Gly Thr Pro Glu Asp
340 345 350

Leu Lys Ser Leu Ile Asp Lys Ala His Glu Leu Gly Leu Leu Val Leu
355 360 365

Met Asp Ile Val His Ser His Ser Ser Asn Asn Thr Leu Asp Gly Leu
370 375 380

Asn Gly Phe Asp Gly Thr Asp Thr His Tyr Phe His Gly Gly Pro Arg
385 390 395 400

Gly His His Trp Met Trp Asp Ser Arg Leu Phe Asn Tyr Gly Ser Trp
405 410 415

Glu Val Leu Arg Phe Leu Leu Ser Asn Ala Arg Trp Trp Leu Glu Glu
420 425 430

Tyr Lys Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser Met Met Tyr
435 440 445

Thr His His Gly Leu Gln Val Thr Phe Thr Gly Asn Tyr Gly Glu Tyr
450 455 460

Phe Gly Phe Ala Thr Asp Val Asp Ala Val Val Tyr Leu Met Leu Val
465 470 475 480

Asn Asp Leu Ile Arg Gly Leu Tyr Pro Glu Ala Val Ser Ile Gly Glu
485 490 495

Asp Val Ser Gly Met Pro Thr Phe Cys Ile Pro Val Gln Asp Gly Gly
500 505 510

Val Gly Phe Asp Tyr Arg Leu His Met Ala Val Pro Asp Lys Trp Ile
515 520 525

Glu Leu Leu Lys Gln Ser Asp Glu Tyr Trp Glu Met Gly Asp Ile Val
530 535 540

His Thr Leu Thr Asn Arg Arg Trp Leu Glu Lys Cys Val Thr Tyr Cys
545 550 555 560

Glu Ser His Asp Gln Ala Leu Val Gly Asp Lys Thr Ile Ala Phe Trp
565 570 575

Leu Met Asp Lys Asp Met Tyr Asp Phe Met Ala Leu Asp Arg Pro Ser
580 585 590

Thr Pro Arg Ile Asp Arg Gly Ile Ala Leu His Lys Met Ile Arg Leu
595 600 605

Val Thr Met Gly Leu Gly Gly Glu Gly Tyr Leu Asn Phe Met Gly Asn
610 615 620

Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Gly Pro Gln Ser
625 630 635 640

Leu Pro Asn Gly Ser Val Ile Pro Gly Asn Asn Asn Ser Phe Asp Lys
645 650 655

Cys Arg Arg Arg Phe Asp Leu Gly Asp Ala Asp Tyr Leu Arg Tyr Arg
660 665 670

Gly Met Gln Glu Phe Asp Gln Ala Met Gln His Leu Glu Gly Lys Tyr
675 680 685

Glu Phe Met Thr Ser Asp His Ser Tyr Val Ser Arg Lys His Glu Glu
690 695 700

Asp Lys Val Ile Ile Phe Glu Arg Gly Asp Leu Val Phe Val Phe Asn
705 710 715 720

Phe His Trp Ser Asn Ser Tyr Phe Asp Tyr Arg Val Gly Cys Phe Lys
725 730 735

Pro Gly Lys Tyr Lys Ile Val Leu Asp Ser Asp Asp Gly Leu Phe Gly
740 745 750

Gly Phe Ser Arg Leu Asp His Asp Ala Glu Tyr Phe Thr Ala Asp Trp
755 760 765

Pro His Asp Asn Arg Pro Cys Ser Phe Ser Val Tyr Ala Pro Ser Arg
770 775 780

Thr Ala Val Val Tyr Ala Pro Ala Gly Ala Glu Asp Glu
785 790 795

<210> 14
<211> 747
<212> PRT
<213> Zea mays

<400> 14
Ala Ala Ala Ala Ala Arg Lys Ala Val Met Val Pro Glu Gly Glu Asn
1 5 10 15

Asp Gly Leu Ala Ser Arg Ala Asp Ser Ala Gln Phe Gln Ser Asp Glu
20 25 30

Leu Glu Val Pro Asp Ile Ser Glu Glu Thr Thr Cys Gly Ala Gly Val
35 40 45

Ala Asp Ala Gln Ala Leu Asn Arg Val Arg Val Val Pro Pro Pro Ser
50 55 60

Asp Gly Gln Lys Ile Phe Gln Ile Asp Pro Met Leu Gln Gly Tyr Lys
65 70 75 80

Tyr His Leu Glu Tyr Arg Tyr Ser Leu Tyr Arg Arg Ile Arg Ser Asp
85 90 95

Ile Asp Glu His Glu Gly Gly Leu Glu Ala Phe Ser Arg Ser Tyr Glu
100 105 110

Lys Phe Gly Phe Asn Ala Ser Ala Glu Gly Ile Thr Tyr Arg Glu Trp
115 120 125

Ala Pro Gly Ala Phe Ser Ala Ala Leu Val Gly Asp Val Asn Asn Trp
130 135 140

Asp Pro Asn Ala Asp Arg Met Ser Lys Asn Glu Phe Gly Val Trp Glu
145 150 155 160

Ile Phe Leu Pro Asn Asn Ala Asp Gly Thr Ser Pro Ile Pro His Gly
165 170 175

Ser Arg Val Lys Val Arg Met Asp Thr Pro Ser Gly Ile Lys Asp Ser
180 185 190

Ile Pro Ala Trp Ile Lys Tyr Ser Val Gln Ala Pro Gly Glu Ile Pro
195 200 205

Tyr Asp Gly Ile Tyr Tyr Asp Pro Pro Glu Glu Val Lys Tyr Val Phe
210 215 220

Arg His Ala Gln Pro Lys Arg Pro Lys Ser Leu Arg Ile Tyr Glu Thr
225 230 235 240

His Val Gly Met Ser Ser Pro Glu Pro Lys Ile Asn Thr Tyr Val Asn
245 250 255

Phe Arg Asp Glu Val Leu Pro Arg Ile Lys Lys Leu Gly Tyr Asn Ala
260 265 270

Val Gln Ile Met Ala Ile Gln Glu His Ser Tyr Tyr Gly Ser Phe Gly
275 280 285

Tyr His Val Thr Asn Phe Phe Ala Pro Ser Ser Arg Phe Gly Thr Pro
290 295 300

Glu Asp Leu Lys Ser Leu Ile Asp Arg Ala His Glu Leu Gly Leu Leu
305 310 315 320

Val Leu Met Asp Val Val His Ser His Ala Ser Ser Asn Thr Leu Asp
325 330 335

Gly Leu Asn Gly Phe Asp Gly Thr Asp Thr His Tyr Phe His Ser Gly
340 345 350

Pro Arg Gly His His Trp Met Trp Asp Ser Arg Leu Phe Asn Tyr Gly
355 360 365

Asn Trp Glu Val Leu Arg Phe Leu Leu Ser Asn Ala Arg Trp Trp Leu
370 375 380

Glu Glu Tyr Lys Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser Met
385 390 395 400

Met Tyr Thr His His Gly Leu Gln Val Thr Phe Thr Gly Asn Phe Asn
405 410 415

Glu Tyr Phe Gly Phe Ala Thr Asp Val Asp Ala Val Val Tyr Leu Met
420 425 430

Leu Val Asn Asp Leu Ile His Gly Leu Tyr Pro Glu Ala Val Thr Ile
435 440 445

Gly Glu Asp Val Ser Gly Met Pro Thr Phe Ala Leu Pro Val His Asp
450 455 460

Gly Gly Val Gly Phe Asp Tyr Arg Met His Met Ala Val Ala Asp Lys
465 470 475 480

Trp Ile Asp Leu Leu Lys Gln Ser Asp Glu Thr Trp Lys Met Gly Asp
485 490 495

Ile Val His Thr Leu Thr Asn Arg Arg Trp Leu Glu Lys Cys Val Thr
500 505 510

Tyr Ala Glu Ser His Asp Gln Ala Leu Val Gly Asp Lys Thr Ile Ala
515 520 525

Phe Trp Leu Met Asp Lys Asp Met Tyr Asp Phe Met Ala Leu Asp Arg
530 535 540

Pro Ser Thr Pro Thr Ile Asp Arg Gly Ile Ala Leu His Lys Met Ile
545 550 555 560

Arg Leu Ile Thr Met Gly Leu Gly Gly Glu Gly Tyr Leu Asn Phe Met
565 570 575

Gly Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Gly Pro
580 585 590

Gln Arg Leu Pro Ser Gly Lys Phe Ile Pro Gly Asn Asn Asn Ser Tyr
595 600 605

Asp Lys Cys Arg Arg Arg Phe Asp Leu Gly Asp Ala Asp Tyr Leu Arg
610 615 620

Tyr His Gly Met Gln Glu Phe Asp Gln Ala Met Gln His Leu Glu Gln
625 630 635 640

Lys Tyr Glu Phe Met Thr Ser Asp His Gln Tyr Ile Ser Arg Lys His
645 650 655

Glu Glu Asp Lys Val Ile Val Phe Glu Lys Gly Asp Leu Val Phe Val
660 665 670

Phe Asn Phe His Cys Asn Asn Ser Tyr Phe Asp Tyr Arg Ile Gly Cys
675 680 685

Arg Lys Pro Gly Val Tyr Lys Val Val Leu Asp Ser Asp Ala Gly Leu
690 695 700

Phe Gly Gly Phe Ser Arg Ile His His Ala Ala Glu His Phe Thr Ala
705 710 715 720

Asp Cys Ser His Asp Asn Arg Pro Tyr Ser Phe Ser Val Tyr Thr Pro
725 730 735

Ser Arg Thr Cys Val Val Tyr Ala Pro Val Glu
740 745

<210> 15
<211> 50
<212> PRT
<213> Hordeum vulgare

<400> 15
Asn Asp Leu Gly Ile Trp Glu Ile Phe Leu Pro Asn Asn Ala Asp Gly
1 5 10 15

Ser Pro Pro Ile Pro His Gly Ser Arg Val Lys Val Arg Met Asp Thr
20 25 30

Pro Ser Gly Thr Lys Asp Ser Ile Pro Ala Trp Ile Lys Phe Ser Val
35 40 45

Gln Ala
50

<210> 16
<211> 50
<212> PRT
<213> Hordeum vulgare

<400> 16
Asp Asp Tyr Gly Val Trp Glu Ile Phe Leu Pro Asn Asn Ala Asp Gly
1 5 10 15

Ser Pro Ala Ile Pro His Gly Ser Arg Val Lys Ile Arg Met Asp Thr
20 25 30

Pro Ser Gly Val Lys Asp Ser Ile Ser Ala Trp Ile Lys Phe Ser Val
35 40 45

Gln Ala
50

<210> 17
<211> 760
<212> PRT
<213> Oryza sativa

<400> 17
Ala Ala Gly Ala Ser Gly Glu Val Met Ile Pro Glu Gly Glu Ser Asp
1 5 10 15

Gly Met Pro Val Ser Ala Gly Ser Asp Asp Leu Gln Leu Pro Ala Leu
20 25 30

Asp Asp Glu Leu Ser Thr Glu Val Gly Ala Glu Val Glu Ile Glu Ser
35 40 45

Ser Gly Ala Ser Asp Val Glu Gly Val Lys Arg Val Val Glu Glu Leu
50 55 60

Ala Ala Glu Gln Lys Pro Arg Val Val Pro Pro Thr Gly Asp Gly Gln
65 70 75 80

Lys Ile Phe Gln Met Asp Ser Met Leu Asn Gly Tyr Lys Tyr His Leu
85 90 95

Glu Tyr Arg Tyr Ser Leu Tyr Arg Arg Leu Arg Ser Asp Ile Asp Gln
100 105 110

Tyr Glu Gly Gly Leu Glu Thr Phe Ser Arg Gly Tyr Glu Lys Phe Gly
115 120 125

```
Phe Asn His Ser Ala Glu Gly Val Thr Tyr Arg Glu Trp Ala Pro Gly
    130                 135                 140

Ala His Ser Ala Ala Leu Val Gly Asp Phe Asn Asn Trp Asn Pro Asn
145                 150                 155                 160

Ala Asp Arg Met Ser Lys Asn Glu Phe Gly Val Trp Glu Ile Phe Leu
                165                 170                 175

Pro Asn Asn Ala Asp Gly Ser Ser Pro Ile Pro His Gly Ser Arg Val
            180                 185                 190

Lys Val Arg Met Glu Thr Pro Ser Gly Ile Lys Asp Ser Ile Pro Ala
        195                 200                 205

Trp Ile Lys Tyr Ser Val Gln Ala Ala Gly Glu Ile Pro Tyr Asn Gly
    210                 215                 220

Ile Tyr Tyr Asp Pro Pro Glu Glu Glu Lys Tyr Ile Phe Lys His Pro
225                 230                 235                 240

Gln Pro Lys Arg Pro Lys Ser Leu Arg Ile Tyr Glu Thr His Val Gly
                245                 250                 255

Met Ser Ser Thr Glu Pro Lys Ile Asn Thr Tyr Ala Asn Phe Arg Asp
            260                 265                 270

Glu Val Leu Pro Arg Ile Lys Lys Leu Gly Tyr Asn Ala Val Gln Ile
        275                 280                 285

Met Ala Ile Gln Glu His Ala Tyr Tyr Gly Ser Phe Gly Tyr His Val
    290                 295                 300

Thr Asn Phe Phe Ala Pro Ser Ser Arg Phe Gly Thr Pro Glu Asp Leu
305                 310                 315                 320

Lys Ser Leu Ile Asp Lys Ala His Glu Leu Gly Leu Val Val Leu Met
                325                 330                 335

Asp Val Val His Ser His Ala Ser Asn Asn Thr Leu Asp Gly Leu Asn
            340                 345                 350

Gly Phe Asp Gly Thr Asp Thr His Tyr Phe His Ser Gly Ser Arg Gly
        355                 360                 365

His His Trp Met Trp Asp Ser Arg Leu Phe Asn Tyr Gly Asn Trp Glu
    370                 375                 380
```

Val Leu Arg Phe Leu Leu Ser Asn Ala Arg Trp Trp Leu Glu Glu Tyr
385 390 395 400

Lys Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser Met Met Tyr Thr
405 410 415

His His Gly Leu Gln Val Ala Phe Thr Gly Asn Tyr Ser Glu Tyr Phe
420 425 430

Gly Phe Ala Thr Asp Ala Asp Ala Val Val Tyr Leu Met Leu Val Asn
435 440 445

Asp Leu Ile His Gly Leu Tyr Pro Glu Ala Ile Thr Ile Gly Glu Asp
450 455 460

Val Ser Gly Met Pro Thr Phe Ala Leu Pro Val Gln Asp Gly Gly Val
465 470 475 480

Gly Phe Asp Tyr Arg Leu His Met Ala Val Pro Asp Lys Trp Ile Glu
485 490 495

Leu Leu Lys Gln Ser Asp Glu Ser Trp Lys Met Gly Asp Ile Val His
500 505 510

Thr Leu Thr Asn Arg Arg Trp Ser Glu Lys Cys Val Thr Tyr Ala Glu
515 520 525

Ser His Asp Gln Ala Leu Val Gly Asp Lys Thr Ile Ala Phe Trp Leu
530 535 540

Met Asp Lys Asp Met Tyr Asp Phe Met Ala Leu Asp Arg Pro Ala Thr
545 550 555 560

Pro Ser Ile Asp Arg Gly Ile Ala Leu His Lys Met Ile Arg Leu Ile
565 570 575

Thr Met Gly Leu Gly Gly Glu Gly Tyr Leu Asn Phe Met Gly Asn Glu
580 585 590

Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Ala Pro Gln Val Leu
595 600 605

Pro Asn Gly Lys Phe Ile Pro Gly Asn Asn Asn Ser Tyr Asp Lys Cys
610 615 620

Arg Arg Arg Phe Asp Leu Gly Asp Ala Asp Tyr Leu Arg Tyr Arg Gly
625 630 635 640

Met Leu Glu Phe Asp Arg Ala Met Gln Ser Leu Glu Glu Lys Tyr Gly
645 650 655

Phe Met Thr Ser Asp His Gln Tyr Ile Ser Arg Lys His Glu Glu Asp
660 665 670

Lys Met Ile Ile Phe Glu Lys Gly Asp Leu Val Phe Val Phe Asn Phe
675 680 685

His Trp Ser Asn Ser Tyr Phe Asp Tyr Arg Val Gly Cys Leu Lys Pro
690 695 700

Gly Lys Tyr Lys Val Val Leu Asp Ser Asp Ala Gly Leu Phe Gly Gly
705 710 715 720

Phe Gly Arg Ile His His Thr Ala Glu His Phe Thr Ala Asp Cys Ser
725 730 735

His Asp Asn Arg Pro Tyr Ser Phe Ser Val Tyr Ser Pro Ser Arg Thr
740 745 750

Cys Val Val Tyr Ala Pro Ala Glu
755 760

<210> 18
<211> 844
<212> PRT
<213> Oryza sativa

<400> 18
Val Glu Ala Glu Arg Gly Gly Cys Arg Gly Ile Arg Ser Gly Cys Gly
1 5 10 15

Ala Gly Glu Met Ala Ala Pro Ala Ser Ala Val Pro Gly Ser Ala Ala
20 25 30

Gly Leu Arg Ala Gly Ala Val Arg Phe Pro Val Pro Ala Gly Ala Arg
35 40 45

Ser Trp Arg Ala Ala Ala Glu Leu Pro Thr Ser Arg Ser Leu Leu Ser
50 55 60

Gly Arg Arg Phe Pro Gly Ala Val Arg Val Gly Gly Ser Gly Gly Arg
65 70 75 80

Val Ala Val Arg Ala Ala Gly Ala Ser Gly Glu Val Met Ile Pro Glu

```
                85                  90                  95

Gly Glu Ser Asp Gly Met Pro Val Ser Ala Gly Ser Asp Asp Leu Gln
            100                 105                 110

Leu Pro Ala Leu Asp Asp Glu Leu Ser Thr Glu Val Gly Ala Glu Val
        115                 120                 125

Glu Ile Glu Ser Ser Gly Ala Ser Asp Val Glu Gly Val Lys Arg Val
    130                 135                 140

Val Glu Glu Leu Ala Ala Glu Gln Lys Pro Arg Val Val Pro Pro Thr
145                 150                 155                 160

Gly Asp Gly Gln Lys Ile Phe Gln Met Asp Ser Met Leu Asn Gly Tyr
                165                 170                 175

Lys Tyr His Leu Glu Tyr Arg Tyr Ser Leu Tyr Arg Arg Leu Arg Ser
            180                 185                 190

Asp Ile Asp Gln Tyr Glu Gly Gly Leu Glu Thr Phe Ser Arg Gly Tyr
        195                 200                 205

Glu Lys Phe Gly Phe Asn His Ser Ala Glu Gly Val Thr Tyr Arg Glu
    210                 215                 220

Trp Ala Pro Gly Ala His Ser Ala Ala Leu Val Gly Asp Phe Asn Asn
225                 230                 235                 240

Trp Asn Pro Asn Ala Asp Arg Met Ser Lys Asn Glu Phe Gly Val Trp
                245                 250                 255

Glu Ile Phe Leu Pro Asn Asn Ala Asp Gly Ser Ser Pro Ile Pro His
            260                 265                 270

Gly Ser Arg Val Lys Val Arg Met Glu Thr Pro Ser Gly Ile Lys Asp
        275                 280                 285

Ser Ile Pro Ala Trp Ile Lys Tyr Ser Val Gln Ala Ala Gly Glu Ile
    290                 295                 300

Pro Tyr Asn Gly Ile Tyr Tyr Asp Pro Pro Glu Glu Glu Lys Tyr Ile
305                 310                 315                 320

Phe Lys His Pro Gln Pro Lys Arg Pro Lys Ser Leu Arg Ile Tyr Glu
                325                 330                 335

Thr His Val Gly Met Ser Ser Thr Glu Pro Lys Ile Asn Thr Tyr Ala
```

340 345 350

Asn Phe Arg Asp Glu Val Leu Pro Arg Ile Lys Lys Leu Gly Tyr Asn
355 360 365

Ala Val Gln Ile Met Ala Ile Gln Glu His Ala Tyr Tyr Gly Ser Phe
370 375 380

Gly Tyr His Val Thr Asn Phe Phe Ala Pro Ser Ser Arg Phe Gly Thr
385 390 395 400

Pro Glu Asp Leu Lys Ser Leu Ile Asp Lys Ala His Glu Leu Gly Leu
405 410 415

Val Val Leu Met Asp Val Val His Ser His Ala Ser Asn Asn Thr Leu
420 425 430

Asp Gly Leu Asn Gly Phe Asp Gly Thr Asp Thr His Tyr Phe His Ser
435 440 445

Gly Ser Arg Gly His His Trp Met Trp Asp Ser Arg Leu Phe Asn Tyr
450 455 460

Gly Asn Trp Glu Val Leu Arg Phe Leu Leu Ser Asn Ala Arg Trp Trp
465 470 475 480

Leu Glu Glu Tyr Lys Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser
485 490 495

Met Met Tyr Thr His His Gly Leu Gln Val Ala Phe Thr Gly Asn Tyr
500 505 510

Ser Glu Tyr Phe Gly Phe Ala Thr Asp Ala Asp Ala Val Val Tyr Leu
515 520 525

Met Leu Val Asn Asp Leu Ile His Gly Leu Tyr Pro Glu Ala Ile Thr
530 535 540

Ile Gly Glu Asp Val Ser Gly Met Pro Thr Phe Ala Leu Pro Val Gln
545 550 555 560

Asp Gly Gly Val Gly Phe Asp Tyr Arg Leu His Met Ala Val Pro Asp
565 570 575

Lys Trp Ile Glu Leu Leu Lys Gln Ser Asp Glu Ser Trp Lys Met Gly
580 585 590

Asp Ile Val His Thr Leu Thr Asn Arg Arg Trp Ser Glu Lys Cys Val

```
        595                     600                     605

Thr Tyr Ala Glu Ser His Asp Gln Ala Leu Val Gly Asp Lys Thr Ile
    610                     615                     620

Ala Phe Trp Leu Met Asp Lys Asp Met Tyr Asp Phe Met Ala Leu Asp
625                     630                     635                     640

Arg Pro Ala Thr Pro Ser Ile Asp Arg Gly Ile Ala Leu His Lys Met
                645                     650                     655

Ile Arg Leu Ile Thr Met Gly Leu Gly Gly Glu Gly Tyr Leu Asn Phe
            660                     665                     670

Met Gly Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Ala
        675                     680                     685

Pro Gln Val Leu Pro Asn Gly Lys Phe Ile Pro Gly Asn Asn Asn Ser
    690                     695                     700

Tyr Asp Lys Cys Arg Arg Arg Phe Asp Leu Gly Asp Ala Asp Tyr Leu
705                     710                     715                     720

Arg Tyr Arg Gly Met Leu Glu Phe Asp Arg Ala Met Gln Ser Leu Glu
                725                     730                     735

Glu Lys Tyr Gly Phe Met Thr Ser Asp His Gln Tyr Ile Ser Arg Lys
            740                     745                     750

His Glu Glu Asp Lys Met Ile Ile Phe Glu Lys Gly Asp Leu Val Phe
        755                     760                     765

Val Phe Asn Phe His Trp Ser Asn Ser Tyr Phe Asp Tyr Arg Val Gly
    770                     775                     780

Cys Leu Lys Pro Gly Lys Tyr Lys Val Val Leu Asp Ser Asp Ala Gly
785                     790                     795                     800

Leu Phe Gly Gly Phe Gly Arg Ile His His Thr Ala Glu His Phe Thr
                805                     810                     815

Ala Asp Cys Ser His Asp Asn Arg Pro Tyr Ser Phe Ser Val Tyr Ser
            820                     825                     830

Pro Ser Arg Thr Cys Val Val Tyr Ala Pro Ala Glu
        835                     840
```

<210> 19
<211> 857
<212> PRT
<213> Pisum sativum

<400> 19

Lys Val Leu Ile Pro Glu Asp Gln Asp Asn Ser Val Ser Leu Ala Asp
1 5 10 15

Gln Leu Glu Asn Pro Asp Ile Thr Ser Glu Asp Ala Gln Asn Leu Glu
20 25 30

Asp Leu Thr Met Lys Asp Gly Asn Lys Tyr Asn Ile Asp Glu Ser Thr
35 40 45

Ser Ser Tyr Arg Glu Val Gly Asp Glu Lys Gly Ser Val Thr Ser Ser
50 55 60

Ser Leu Val Asp Val Asn Thr Asp Thr Gln Ala Lys Lys Thr Ser Val
65 70 75 80

His Ser Asp Lys Lys Val Lys Val Asp Lys Pro Lys Ile Ile Pro Pro
85 90 95

Pro Gly Thr Gly Gln Lys Ile Tyr Glu Ile Asp Pro Leu Leu Gln Ala
100 105 110

His Arg Gln His Leu Asp Phe Arg Tyr Gly Gln Tyr Lys Arg Ile Arg
115 120 125

Glu Glu Ile Asp Lys Tyr Glu Gly Gly Leu Asp Ala Phe Ser Arg Gly
130 135 140

Tyr Glu Lys Phe Gly Phe Thr Arg Ser Ala Thr Gly Ile Thr Tyr Arg
145 150 155 160

Glu Trp Ala Pro Gly Ala Lys Ser Ala Ala Leu Val Gly Asp Phe Asn
165 170 175

Asn Trp Asn Pro Asn Ala Asp Val Met Thr Lys Asp Ala Phe Gly Val
180 185 190

Trp Glu Ile Phe Leu Pro Asn Asn Ala Asp Gly Ser Pro Pro Ile Pro
195 200 205

His Gly Ser Arg Val Lys Ile His Met Asp Thr Pro Ser Gly Ile Lys
210 215 220

Asp Ser Ile Pro Ala Trp Ile Lys Phe Ser Val Gln Ala Pro Gly Glu
225 230 235 240

Ile Pro Tyr Asn Gly Ile Tyr Tyr Asp Pro Pro Glu Glu Glu Lys Tyr
245 250 255

Val Phe Lys His Pro Gln Pro Lys Arg Pro Gln Ser Ile Arg Ile Tyr
260 265 270

Glu Ser His Ile Gly Met Ser Ser Pro Glu Pro Lys Ile Asn Thr Tyr
275 280 285

Ala Asn Phe Arg Asp Asp Val Leu Pro Arg Ile Lys Lys Leu Gly Tyr
290 295 300

Asn Ala Val Gln Ile Met Ala Ile Gln Glu His Ser Tyr Tyr Ala Ser
305 310 315 320

Phe Gly Tyr His Val Thr Asn Phe Phe Ala Pro Ser Ser Arg Phe Gly
325 330 335

Thr Pro Glu Asp Leu Lys Ser Leu Ile Asp Arg Ala His Glu Leu Gly
340 345 350

Leu Leu Val Leu Met Asp Ile Val His Ser His Ser Ser Asn Asn Thr
355 360 365

Leu Asp Gly Leu Asn Met Phe Asp Gly Thr Asp Gly His Tyr Phe His
370 375 380

Pro Gly Ser Arg Gly Tyr His Trp Met Trp Asp Ser Arg Leu Phe Asn
385 390 395 400

Tyr Gly Ser Trp Glu Val Leu Arg Tyr Leu Leu Ser Asn Ala Arg Trp
405 410 415

Trp Leu Asp Glu Tyr Lys Phe Asp Gly Phe Arg Phe Asp Gly Val Thr
420 425 430

Ser Met Met Tyr Thr His His Gly Leu Gln Val Ser Phe Thr Gly Asn
435 440 445

Tyr Ser Glu Tyr Phe Gly Leu Ala Thr Asp Val Glu Ala Val Val Tyr
450 455 460

Met Met Leu Val Asn Asp Leu Ile His Gly Leu Phe Pro Glu Ala Val
465 470 475 480

```
Ser Ile Gly Glu Asp Val Ser Gly Met Pro Thr Phe Cys Leu Pro Thr
                485                 490                 495

Gln Asp Gly Gly Ile Gly Phe Asn Tyr Arg Leu His Met Ala Val Ala
            500                 505                 510

Asp Lys Trp Ile Glu Leu Leu Lys Lys Gln Asp Glu Asp Trp Arg Met
        515                 520                 525

Gly Asp Ile Val His Thr Leu Thr Asn Arg Arg Trp Leu Glu Lys Cys
    530                 535                 540

Val Val Tyr Ala Glu Ser His Asp Gln Ala Leu Val Gly Asp Lys Thr
545                 550                 555                 560

Leu Ala Phe Trp Leu Met Asp Lys Asp Met Tyr Asp Phe Met Ala Leu
                565                 570                 575

Asp Arg Pro Ser Thr Pro Leu Ile Asp Arg Gly Ile Ala Leu His Lys
            580                 585                 590

Met Ile Arg Leu Ile Thr Met Gly Leu Gly Gly Glu Gly Tyr Leu Asn
        595                 600                 605

Phe Met Gly Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg
    610                 615                 620

Gly Glu Gln His Leu Pro Asn Gly Lys Ile Val Pro Gly Asn Asn Asn
625                 630                 635                 640

Ser Tyr Asp Lys Cys Arg Arg Arg Phe Asp Leu Gly Asp Ala Asp Tyr
                645                 650                 655

Leu Arg Tyr His Gly Met Gln Glu Phe Asp Arg Ala Met Gln His Leu
            660                 665                 670

Glu Glu Arg Tyr Gly Phe Met Thr Ser Glu His Gln Tyr Ile Ser Arg
        675                 680                 685

Lys Asn Glu Gly Asp Arg Val Ile Ile Phe Glu Arg Asp Asn Leu Val
    690                 695                 700

Phe Val Phe Asn Phe His Trp Thr Asn Ser Tyr Ser Asp Tyr Lys Val
705                 710                 715                 720

Gly Cys Leu Lys Pro Gly Lys Tyr Lys Ile Val Leu Asp Ser Asp Asp
                725                 730                 735
```

Thr Leu Phe Gly Gly Phe Asn Arg Leu Asn His Thr Ala Glu Tyr Phe
740 745 750

Thr Ser Glu Gly Trp Tyr Asp Asp Arg Pro Arg Ser Phe Leu Val Tyr
755 760 765

Ala Pro Ser Arg Thr Ala Val Val Tyr Ala Leu Ala Asp Gly Val Glu
770 775 780

Ser Glu Pro Ile Glu Leu Ser Asp Gly Val Glu Ser Glu Pro Ile Glu
785 790 795 800

Leu Ser Val Gly Val Glu Ser Glu Pro Ile Glu Leu Ser Val Glu Glu
805 810 815

Ala Glu Ser Glu Pro Ile Glu Arg Ser Val Glu Glu Val Glu Ser Glu
820 825 830

Thr Thr Gln Gln Ser Val Glu Val Glu Ser Glu Thr Thr Gln Gln Ser
835 840 845

Val Glu Val Glu Ser Glu Thr Thr Gln
850 855

<210> 20
<211> 779
<212> PRT
<213> Solanum tuberosum

<400> 20
Thr Met Ala Pro Leu Glu Glu Asp Val Lys Thr Glu Asn Ile Gly Leu
1 5 10 15

Leu Asn Leu Asp Pro Thr Leu Glu Pro Tyr Leu Asp His Phe Arg His
20 25 30

Arg Met Lys Arg Tyr Val Asp Gln Lys Met Leu Ile Glu Lys Tyr Glu
35 40 45

Gly Pro Leu Glu Glu Phe Ala Gln Gly Tyr Leu Lys Phe Gly Phe Asn
50 55 60

Arg Glu Asp Gly Cys Ile Val Tyr Arg Glu Trp Ala Pro Ala Ala Gln
65 70 75 80

Glu Asp Glu Val Ile Gly Asp Phe Asn Gly Trp Asn Gly Ser Asn His
85 90 95

Met Met Glu Lys Asp Gln Phe Gly Val Trp Ser Ile Arg Ile Pro Asp
100 105 110

Val Asp Ser Lys Pro Val Ile Pro His Asn Ser Arg Val Lys Phe Arg
115 120 125

Phe Lys His Gly Asn Gly Val Trp Val Asp Arg Ile Pro Ala Trp Ile
130 135 140

Lys Tyr Ala Thr Ala Asp Ala Thr Lys Phe Ala Ala Pro Tyr Asp Gly
145 150 155 160

Val Tyr Trp Asp Pro Pro Pro Ser Glu Arg Tyr His Phe Lys Tyr Pro
165 170 175

Arg Pro Pro Lys Pro Arg Ala Pro Arg Ile Tyr Glu Ala His Val Gly
180 185 190

Met Ser Ser Ser Glu Pro Arg Val Asn Ser Tyr Arg Glu Phe Ala Asp
195 200 205

Asp Val Leu Pro Arg Ile Lys Ala Asn Asn Tyr Asn Thr Val Gln Leu
210 215 220

Met Ala Ile Met Glu His Ser Tyr Tyr Gly Ser Phe Gly Tyr His Val
225 230 235 240

Thr Asn Phe Phe Ala Val Ser Ser Arg Tyr Gly Asn Pro Glu Asp Leu
245 250 255

Lys Tyr Leu Ile Asp Lys Ala His Ser Leu Gly Leu Gln Val Leu Val
260 265 270

Asp Val Val His Ser His Ala Ser Asn Asn Val Thr Asp Gly Leu Asn
275 280 285

Gly Phe Asp Ile Gly Gln Gly Ser Gln Glu Ser Tyr Phe His Ala Gly
290 295 300

Glu Arg Gly Tyr His Lys Leu Trp Asp Ser Arg Leu Phe Asn Tyr Ala
305 310 315 320

Asn Trp Glu Val Leu Arg Phe Leu Leu Ser Asn Leu Arg Trp Trp Leu
325 330 335

Glu Glu Tyr Asn Phe Asp Gly Phe Arg Phe Asp Gly Ile Thr Ser Met
340 345 350

Leu Tyr Val His His Gly Ile Asn Met Gly Phe Thr Gly Asn Tyr Asn
355 360 365

Glu Tyr Phe Ser Glu Ala Thr Asp Val Asp Ala Val Val Tyr Leu Met
370 375 380

Leu Ala Asn Asn Leu Ile His Lys Ile Phe Pro Asp Ala Thr Val Ile
385 390 395 400

Ala Glu Asp Val Ser Gly Met Pro Gly Leu Gly Arg Pro Val Ser Glu
405 410 415

Gly Gly Ile Gly Phe Asp Tyr Arg Leu Ala Met Ala Ile Pro Asp Lys
420 425 430

Trp Ile Asp Tyr Leu Lys Asn Lys Asn Asp Glu Asp Trp Ser Met Lys
435 440 445

Glu Val Thr Ser Ser Leu Thr Asn Arg Arg Tyr Thr Glu Lys Cys Ile
450 455 460

Ala Tyr Ala Glu Ser His Asp Gln Ser Ile Val Gly Asp Lys Thr Ile
465 470 475 480

Ala Phe Leu Leu Met Asp Lys Glu Met Tyr Ser Gly Met Ser Cys Leu
485 490 495

Thr Asp Ala Ser Pro Val Val Asp Arg Gly Ile Ala Leu His Lys Met
500 505 510

Ile His Phe Phe Thr Met Ala Leu Gly Gly Glu Gly Tyr Leu Asn Phe
515 520 525

Met Gly Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Glu
530 535 540

Gly Asn Asn Trp Ser Tyr Asp Lys Cys Arg Arg Gln Trp Asn Leu Ala
545 550 555 560

Asp Ser Glu His Leu Arg Tyr Lys Phe Met Asn Ala Phe Asp Arg Ala
565 570 575

Met Asn Ser Leu Asp Glu Lys Phe Ser Phe Leu Ala Ser Gly Lys Gln
580 585 590

Ile Val Ser Ser Met Asp Asp Asp Asn Lys Val Val Val Phe Glu Arg
595 600 605

Gly Asp Leu Val Phe Val Phe Asn Phe His Pro Lys Asn Thr Tyr Glu
    610   615   620

Gly Tyr Lys Val Gly Cys Asp Leu Pro Gly Lys Tyr Arg Val Ala Leu
625   630   635   640

Asp Ser Asp Ala Trp Glu Phe Gly Gly His Gly Arg Thr Gly His Asp
                645   650   655

Val Asp His Phe Thr Ser Pro Glu Gly Ile Pro Gly Val Pro Glu Thr
            660   665   670

Asn Phe Asn Gly Arg Gln Ile Pro Ser Lys Cys Cys Leu Leu Arg Glu
        675   680   685

His Val Trp Leu Ile Thr Glu Leu Met Asn Ala Cys Gln Lys Leu Lys
    690   695   700

Ile Thr Arg Gln Thr Phe Val Val Ser Tyr Tyr Gln Gln Pro Ile Ser
705   710   715   720

Arg Arg Val Thr Arg Asn Leu Lys Ile Arg Tyr Leu Gln Ile Ser Val
                725   730   735

Thr Leu Thr Asn Ala Cys Gln Lys Leu Lys Phe Thr Arg Gln Thr Phe
            740   745   750

Leu Val Ser Tyr Tyr Gln Gln Pro Ile Leu Arg Arg Val Thr Arg Lys
        755   760   765

Leu Lys Asp Ser Leu Ser Thr Asn Ile Ser Thr
    770   775

<210> 21
<211> 762
<212> PRT
<213> Triticum aestivum

<400> 21
Thr Met Ala Thr Ala Glu Asp Gly Val Gly Asp Leu Pro Ile Tyr Asp
  1   5   10   15

Leu Asp Pro Lys Phe Ala Gly Phe Lys Glu His Phe Ser Tyr Arg Met
            20   25   30

```
Lys Lys Tyr Leu Asp Gln Lys His Ser Ile Glu Lys His Glu Gly Gly
         35                  40                  45

Leu Glu Glu Phe Ser Lys Gly Tyr Leu Lys Phe Gly Ile Asn Thr Glu
     50                  55                  60

Asn Asp Ala Thr Val Tyr Arg Glu Trp Ala Pro Ala Ala Met Asp Ala
 65                  70                  75                  80

Gln Leu Ile Gly Asp Phe Asn Asn Trp Asn Gly Ser Gly His Arg Met
                 85                  90                  95

Thr Lys Asp Asn Tyr Gly Val Trp Ser Ile Arg Ile Ser His Val Asn
            100                 105                 110

Gly Lys Pro Ala Ile Pro His Asn Ser Lys Val Lys Phe Arg Phe His
        115                 120                 125

Arg Gly Asp Gly Leu Trp Val Asp Arg Val Pro Ala Trp Ile Arg Tyr
    130                 135                 140

Ala Thr Phe Asp Ala Ser Lys Phe Gly Ala Pro Tyr Asp Gly Val His
145                 150                 155                 160

Trp Asp Pro Pro Ser Gly Glu Arg Tyr Val Phe Lys His Pro Arg Pro
                165                 170                 175

Arg Lys Pro Asp Ala Pro Arg Ile Tyr Glu Ala His Val Gly Met Ser
            180                 185                 190

Gly Glu Lys Pro Glu Val Ser Thr Tyr Arg Glu Phe Ala Asp Asn Val
        195                 200                 205

Leu Pro Arg Ile Lys Ala Asn Asn Tyr Asn Thr Val Gln Leu Met Ala
    210                 215                 220

Ile Met Glu His Ser Tyr Tyr Ala Ser Phe Gly Tyr His Val Thr Asn
225                 230                 235                 240

Phe Phe Ala Val Ser Ser Arg Ser Gly Thr Pro Glu Asp Leu Lys Tyr
                245                 250                 255

Leu Val Asp Lys Ala His Ser Leu Gly Leu Arg Val Leu Met Asp Val
            260                 265                 270

Val His Ser His Ala Ser Ser Asn Lys Thr Asp Gly Leu Asn Gly Tyr
        275                 280                 285
```

Asp Val Gly Gln Asn Thr Gln Glu Ser Tyr Phe His Thr Gly Glu Arg
290 295 300

Gly Tyr His Lys Leu Trp Asp Ser Arg Leu Phe Asn Tyr Ala Asn Trp
305 310 315 320

Glu Val Leu Arg Phe Leu Leu Ser Asn Leu Arg Tyr Trp Met Asp Glu
325 330 335

Phe Met Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser Met Leu Tyr
340 345 350

Asn His His Gly Ile Asn Met Ser Phe Ala Gly Ser Tyr Lys Glu Tyr
355 360 365

Phe Gly Leu Asp Thr Asp Val Asp Ala Val Val Tyr Leu Met Leu Ala
370 375 380

Asn His Leu Met His Lys Leu Leu Pro Glu Ala Thr Val Val Ala Glu
385 390 395 400

Asp Val Ser Gly Met Pro Val Leu Cys Arg Ser Val Asp Glu Gly Gly
405 410 415

Val Gly Phe Asp Tyr Arg Leu Ala Met Ala Ile Pro Asp Arg Trp Ile
420 425 430

Asp Tyr Leu Lys Asn Lys Asp Asp Leu Glu Trp Ser Met Ser Gly Ile
435 440 445

Ala His Thr Leu Thr Asn Arg Arg Tyr Thr Glu Lys Cys Ile Ala Tyr
450 455 460

Ala Glu Ser His Asp Gln Ser Ile Val Gly Asp Lys Thr Met Ala Phe
465 470 475 480

Leu Leu Met Asp Lys Glu Met Tyr Thr Gly Met Ser Asp Leu Gln Pro
485 490 495

Ala Ser Pro Thr Ile Asp Arg Gly Ile Ala Leu Gln Lys Met Ile His
500 505 510

Phe Ile Thr Met Ala Leu Gly Gly Asp Gly Tyr Leu Asn Phe Met Gly
515 520 525

Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Glu Gly Asn
530 535 540

Asn Trp Ser Tyr Asp Lys Cys Arg Arg Gln Trp Ser Leu Ala Asp Ile
545 550 555 560

Asp His Leu Arg Tyr Lys Tyr Met Asn Ala Phe Asp Gln Ala Met Asn
565 570 575

Ala Leu Asp Asp Lys Phe Ser Phe Leu Ser Ser Ser Lys Gln Ile Val
580 585 590

Ser Asp Met Asn Glu Glu Lys Lys Ile Ile Val Phe Glu Arg Gly Asp
595 600 605

Leu Val Phe Val Phe Asn Phe His Pro Ser Lys Thr Tyr Asp Gly Tyr
610 615 620

Lys Val Gly Cys Asp Leu Pro Gly Lys Tyr Lys Val Ala Leu Asp Ser
625 630 635 640

Asp Ala Leu Met Phe Gly Gly His Gly Arg Val Ala His Asp Asn Asp
645 650 655

His Phe Thr Ser Pro Glu Gly Val Pro Gly Val Pro Glu Thr Asn Phe
660 665 670

Asn Asn Arg Pro Asn Ser Phe Lys Ile Leu Ser Pro Ser Arg Thr Cys
675 680 685

Val Ala Tyr Tyr Arg Val Glu Glu Lys Ala Glu Lys Pro Lys Asp Glu
690 695 700

Gly Ala Ala Ser Trp Gly Lys Thr Ala Leu Gly Tyr Ile Asp Val Glu
705 710 715 720

Ala Thr Gly Val Lys Asp Ala Ala Asp Gly Glu Ala Thr Ser Gly Ser
725 730 735

Glu Lys Ala Ser Thr Gly Gly Asp Ser Ser Lys Lys Gly Ile Asn Phe
740 745 750

Val Phe Leu Ser Pro Asp Lys Asp Asn Lys
755 760

<210> 22
<211> 703
<212> PRT
<213> Triticum aestivum

<400> 22

Ser Pro Pro Thr Leu Thr Ser Pro Pro Pro Ser Ala Val Pro Ser Thr
1 5 10 15

Thr Met Leu Cys Leu Ser Ser Ser Leu Leu Pro Arg Pro Ser Ala Ala
20 25 30

Ala Asp Arg Pro Leu Pro Gly Ile Ile Ala Gly Gly Gly Gly Gly Lys
35 40 45

Arg Leu Ser Val Val Pro Ser Val Pro Phe Leu Leu Arg Trp Leu Trp
50 55 60

Pro Arg Lys Ala Lys Ser Lys Ser Phe Val Ser Val Thr Ala Arg Gly
65 70 75 80

Asn Lys Ile Ala Ala Thr Thr Gly Tyr Gly Ser Asp His Leu Pro Ile
85 90 95

Tyr Asp Leu Asp Leu Lys Leu Ala Glu Phe Lys Asp His Phe Asp Tyr
100 105 110

Thr Arg Asn Arg Tyr Ile Glu Gln Lys His Leu Ile Glu Lys His Glu
115 120 125

Gly Ser Leu Glu Glu Phe Ser Lys Gly Tyr Leu Lys Phe Gly Ile Asn
130 135 140

Thr Glu His Gly Ala Ser Val Tyr Arg Glu Trp Ala Pro Ala Ala Glu
145 150 155 160

Glu Ala Gln Leu Val Gly Asp Phe Asn Asn Trp Asn Gly Ser Gly His
165 170 175

Lys Met Ala Lys Asp Asn Phe Gly Val Trp Ser Ile Arg Ile Ser His
180 185 190

Val Asn Gly Lys Pro Ala Ile Pro His Asn Ser Lys Val Lys Phe Arg
195 200 205

Phe Arg His His Gly Val Trp Val Glu Gln Ile Pro Ala Trp Ile Arg
210 215 220

Tyr Ala Thr Val Thr Ala Ser Glu Ser Gly Ala Pro Tyr Asp Gly Leu
225 230 235 240

His Trp Asp Pro Pro Ser Ser Glu Arg Tyr Val Phe Asn His Pro Arg
245 250 255

```
Pro Pro Lys Pro Asp Val Pro Arg Ile Tyr Glu Ala His Val Gly Val
            260                 265                 270

Ser Gly Gly Lys Leu Glu Ala Gly Thr Tyr Arg Glu Phe Pro Asp Asn
        275                 280                 285

Val Leu Pro Cys Leu Arg Ala Thr Asn Tyr Asn Thr Val Gln Leu Met
    290                 295                 300

Gly Ile Met Glu His Ser Asp Ser Ala Ser Phe Gly Tyr His Val Thr
305                 310                 315                 320

Asn Phe Phe Ala Val Ser Ser Arg Ser Gly Thr Pro Glu Asp Leu Lys
                325                 330                 335

Tyr Leu Ile Asp Lys Ala His Ser Leu Gly Leu Arg Val Leu Met Asp
            340                 345                 350

Val Val His Ser His Ala Ser Asn Asn Val Ile Asp Gly Leu Asn Gly
        355                 360                 365

Tyr Asp Val Gly Gln Ser Ala His Glu Ser Tyr Phe Tyr Thr Gly Asp
    370                 375                 380

Lys Gly Tyr Asn Lys Met Trp Asn Gly Arg Met Phe Asn Tyr Ala Asn
385                 390                 395                 400

Trp Glu Val Leu Arg Phe Leu Leu Ser Asn Leu Arg Tyr Trp Met Asp
                405                 410                 415

Glu Phe Met Phe Asp Gly Phe Arg Phe Val Gly Val Thr Ser Met Leu
            420                 425                 430

Tyr Asn His Asn Gly Ile Asn Met Ser Phe Asn Gly Asn Tyr Lys Asp
        435                 440                 445

Tyr Ile Gly Leu Asp Thr Asn Val Asp Ala Phe Val Tyr Met Met Leu
    450                 455                 460

Ala Asn His Leu Met His Lys Leu Phe Pro Glu Ala Ile Val Val Ala
465                 470                 475                 480

Val Asp Val Ser Gly Met Pro Val Leu Cys Trp Pro Val Asp Glu Gly
                485                 490                 495

Gly Leu Gly Phe Asp Tyr Arg Gln Ala Met Thr Ile Pro Asp Arg Trp
            500                 505                 510
```

Ile Asp Tyr Leu Glu Asn Lys Gly Asp Gln Gln Trp Ser Met Ser Ser
515 520 525

Val Ile Ser Gln Thr Leu Thr Asn Arg Arg Tyr Pro Glu Lys Phe Ile
530 535 540

Ala Tyr Ala Glu Arg Gln Asn His Ser Ile Ile Gly Ser Lys Thr Met
545 550 555 560

Ala Phe Leu Leu Met Glu Trp Glu Thr Tyr Ser Gly Met Ser Ala Met
565 570 575

Asp Pro Asp Ser Pro Thr Ile Asp Arg Ala Ile Ala Leu Gln Lys Met
580 585 590

Ile His Phe Ile Thr Met Ala Phe Gly Gly Asp Ser Tyr Leu Lys Phe
595 600 605

Met Gly Asn Glu Tyr Met Asn Ala Phe Val Gln Ala Val Asp Thr Pro
610 615 620

Ser Asp Lys Cys Ser Phe Leu Ser Ser Ser Asn Gln Thr Ala Ser His
625 630 635 640

Met Asn Glu Glu Glu Lys Gly Ser Ala Leu Thr Lys Gly Tyr Thr His
645 650 655

Leu Arg Ser Gly Cys Phe Asp Pro Ser Leu Pro Ser Thr Ser Ser Cys
660 665 670

Ala Phe Leu Gly Pro Ser Asn Gln Ser Pro Phe Ser Lys Pro Phe Ile
675 680 685

Gly Phe Pro Gly Cys Ile Phe Cys Cys Gly Leu Phe Lys Gly Glu
690 695 700

<210> 23
<211> 752
<212> PRT
<213> Zea mays

<400> 23
Thr Met Ala Thr Ala Lys Gly Asp Val Asp His Leu Pro Ile Tyr Asp
1 5 10 15

Leu Asp Pro Lys Leu Glu Ile Phe Lys Asp His Phe Arg Tyr Arg Met

20 25 30

Lys Arg Phe Leu Glu Gln Lys Gly Ser Ile Glu Glu Asn Glu Gly Ser
35 40 45

Leu Glu Ser Phe Ser Lys Gly Tyr Leu Lys Phe Gly Ile Asn Thr Asn
50 55 60

Glu Asp Gly Thr Val Tyr Arg Glu Trp Ala Pro Ala Ala Gln Glu Ala
65 70 75 80

Glu Leu Ile Gly Asp Phe Asn Asp Trp Asn Gly Ala Asn His Lys Met
85 90 95

Glu Lys Asp Lys Phe Gly Val Trp Ser Ile Lys Ile Asp His Val Lys
100 105 110

Gly Lys Pro Ala Ile Pro His Asn Ser Lys Val Lys Phe Arg Phe Leu
115 120 125

His Gly Gly Val Trp Val Asp Arg Ile Pro Ala Leu Ile Arg Tyr Ala
130 135 140

Thr Val Asp Ala Ser Lys Phe Gly Ala Pro Tyr Asp Gly Val His Trp
145 150 155 160

Asp Pro Pro Ala Ser Glu Arg Tyr Thr Phe Lys His Pro Arg Pro Ser
165 170 175

Lys Pro Ala Ala Pro Arg Ile Tyr Glu Ala His Val Gly Met Ser Gly
180 185 190

Glu Lys Pro Ala Val Ser Thr Tyr Arg Glu Phe Ala Asp Asn Val Leu
195 200 205

Pro Arg Ile Arg Ala Asn Asn Tyr Asn Thr Val Gln Leu Met Ala Val
210 215 220

Met Glu His Ser Tyr Tyr Ala Ser Phe Gly Tyr His Val Thr Asn Phe
225 230 235 240

Phe Ala Val Ser Ser Arg Ser Gly Thr Pro Glu Asp Leu Lys Tyr Leu
245 250 255

Val Asp Lys Ala His Ser Leu Gly Leu Arg Val Leu Met Asp Val Val
260 265 270

His Ser His Ala Ser Asn Asn Val Thr Asp Gly Leu Asn Gly Tyr Asp

275 280 285

Val Gly Gln Ser Thr Gln Glu Ser Tyr Phe His Ala Gly Asp Arg Gly
290 295 300

Tyr His Lys Leu Trp Asp Ser Arg Leu Phe Asn Tyr Ala Asn Trp Glu
305 310 315 320

Val Leu Arg Phe Leu Leu Ser Asn Leu Arg Tyr Trp Leu Asp Glu Phe
325 330 335

Met Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser Met Leu Tyr His
340 345 350

His His Gly Ile Asn Val Gly Phe Thr Gly Asn Tyr Gln Glu Tyr Phe
355 360 365

Ser Leu Asp Thr Ala Val Asp Ala Val Val Tyr Met Met Leu Ala Asn
370 375 380

His Leu Met His Lys Leu Leu Pro Glu Ala Thr Val Val Ala Glu Asp
385 390 395 400

Val Ser Gly Met Pro Val Leu Cys Arg Pro Val Asp Glu Gly Gly Val
405 410 415

Gly Phe Asp Tyr Arg Leu Ala Met Ala Ile Pro Asp Arg Trp Ile Asp
420 425 430

Tyr Leu Lys Asn Lys Asp Asp Ser Glu Trp Ser Met Gly Glu Ile Ala
435 440 445

His Thr Leu Thr Asn Arg Arg Tyr Thr Glu Lys Cys Ile Ala Tyr Ala
450 455 460

Glu Ser His Asp Gln Ser Ile Val Gly Asp Lys Thr Ile Ala Phe Leu
465 470 475 480

Leu Met Asp Lys Glu Met Tyr Thr Gly Met Ser Asp Leu Gln Pro Ala
485 490 495

Ser Pro Thr Ile Asp Arg Gly Ile Ala Leu Gln Lys Met Ile His Phe
500 505 510

Ile Thr Met Ala Leu Gly Gly Asp Gly Tyr Leu Asn Phe Met Gly Asn
515 520 525

Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Glu Gly Asn Asn

530 535 540

Trp Ser Tyr Asp Lys Cys Arg Arg Gln Trp Ser Leu Val Asp Thr Asp
545 550 555 560

His Leu Arg Tyr Lys Tyr Met Asn Ala Phe Asp Gln Ala Met Asn Ala
565 570 575

Leu Asp Glu Arg Phe Ser Phe Leu Ser Ser Ser Lys Gln Ile Val Ser
580 585 590

Asp Met Asn Asp Glu Glu Lys Val Ile Val Phe Glu Arg Gly Asp Leu
595 600 605

Val Phe Val Phe Asn Phe His Pro Lys Lys Thr Tyr Glu Gly Tyr Lys
610 615 620

Val Gly Cys Asp Leu Pro Gly Lys Tyr Arg Val Ala Leu Asp Ser Asp
625 630 635 640

Ala Leu Val Phe Gly Gly His Gly Arg Val Gly His Asp Val Asp His
645 650 655

Phe Thr Ser Pro Glu Gly Val Pro Gly Val Pro Glu Thr Asn Phe Asn
660 665 670

Asn Arg Pro Asn Ser Phe Lys Val Leu Ser Pro Pro Arg Thr Cys Val
675 680 685

Ala Tyr Tyr Arg Val Asp Glu Ala Gly Ala Gly Arg Arg Leu His Ala
690 695 700

Lys Ala Glu Thr Gly Lys Thr Ser Pro Ala Glu Ser Ile Asp Val Lys
705 710 715 720

Ala Ser Arg Ala Ser Ser Lys Glu Asp Lys Glu Ala Thr Ala Gly Gly
725 730 735

Lys Lys Gly Trp Lys Phe Ala Arg Gln Pro Ser Asp Gln Asp Thr Lys
740 745 750

<210> 24
<211> 756
<212> PRT

<213> Oryza sativa

<400> 24

Thr Met Val Thr Val Val Glu Glu Val Asp His Leu Pro Ile Tyr Asp
1 5 10 15

Leu Asp Pro Lys Leu Glu Glu Phe Lys Asp His Phe Asn Tyr Arg Ile
20 25 30

Lys Arg Tyr Leu Asp Gln Lys Cys Leu Ile Glu Lys His Glu Gly Gly
35 40 45

Leu Glu Glu Phe Ser Lys Gly Tyr Leu Lys Phe Gly Ile Asn Thr Val
50 55 60

Asp Gly Ala Thr Ile Tyr Arg Glu Trp Ala Pro Ala Ala Gln Glu Ala
65 70 75 80

Gln Leu Ile Gly Glu Phe Asn Asn Trp Asn Gly Ala Lys His Lys Met
85 90 95

Glu Lys Asp Lys Phe Gly Ile Trp Ser Ile Lys Ile Ser His Val Asn
100 105 110

Gly Lys Pro Ala Ile Pro His Asn Ser Lys Val Lys Phe Arg Phe Arg
115 120 125

His Gly Gly Gly Ala Trp Val Asp Arg Ile Pro Ala Trp Ile Arg Tyr
130 135 140

Ala Thr Phe Asp Ala Ser Lys Phe Gly Ala Pro Tyr Asp Gly Val His
145 150 155 160

Trp Asp Pro Pro Ala Cys Glu Arg Tyr Val Phe Lys His Pro Arg Pro
165 170 175

Pro Lys Pro Asp Ala Pro Arg Ile Tyr Glu Ala His Val Gly Met Ser
180 185 190

Gly Glu Glu Pro Glu Val Ser Thr Tyr Arg Glu Phe Ala Asp Asn Val
195 200 205

Leu Pro Arg Ile Arg Ala Asn Asn Tyr Asn Thr Val Gln Leu Met Ala
210 215 220

Ile Met Glu His Ser Tyr Tyr Ala Ser Phe Gly Tyr His Val Thr Asn
225 230 235 240

Phe Phe Ala Val Ser Ser Arg Ser Gly Thr Pro Glu Asp Leu Lys Tyr
245 250 255

Leu Val Asp Lys Ala His Ser Leu Gly Leu Arg Val Leu Met Asp Val
260 265 270

Val His Ser His Ala Ser Asn Asn Val Thr Asp Gly Leu Asn Gly Tyr
275 280 285

Asp Val Gly Gln Asn Thr His Glu Ser Tyr Phe His Thr Gly Asp Arg
290 295 300

Gly Tyr His Lys Leu Trp Asp Ser Arg Leu Phe Asn Tyr Ala Asn Trp
305 310 315 320

Glu Val Leu Arg Phe Leu Leu Ser Asn Leu Arg Tyr Trp Met Asp Glu
325 330 335

Phe Met Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser Met Leu Tyr
340 345 350

His His His Gly Ile Asn Lys Gly Phe Thr Gly Asn Tyr Lys Glu Tyr
355 360 365

Phe Ser Leu Asp Thr Asp Val Asp Ala Ile Val Tyr Met Met Leu Ala
370 375 380

Asn His Leu Met His Lys Leu Leu Pro Glu Ala Thr Ile Val Ala Glu
385 390 395 400

Asp Val Ser Gly Met Pro Val Leu Cys Arg Pro Val Asp Glu Gly Gly
405 410 415

Val Gly Phe Asp Phe Arg Leu Ala Met Ala Ile Pro Asp Arg Trp Ile
420 425 430

Asp Tyr Leu Lys Asn Lys Glu Asp Arg Lys Trp Ser Met Ser Glu Ile
435 440 445

Val Gln Thr Leu Thr Asn Arg Arg Tyr Thr Glu Lys Cys Ile Ala Tyr
450 455 460

Ala Glu Ser His Asp Gln Ser Ile Val Gly Asp Lys Thr Ile Ala Phe
465 470 475 480

Leu Leu Met Asp Lys Glu Met Tyr Thr Gly Met Ser Asp Leu Gln Pro
485 490 495

```
Ala Ser Pro Thr Ile Asn Arg Gly Ile Ala Leu Gln Lys Met Ile His
            500                 505                 510

Phe Ile Thr Met Ala Leu Gly Gly Asp Gly Tyr Leu Asn Phe Met Gly
        515                 520                 525

Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Glu Gly Asn
    530                 535                 540

Asn Trp Ser Tyr Asp Lys Cys Arg Arg Gln Trp Ser Leu Val Asp Thr
545                 550                 555                 560

Asp His Leu Arg Tyr Lys Tyr Met Asn Ala Phe Asp Gln Ala Met Asn
                565                 570                 575

Ala Leu Glu Glu Glu Phe Ser Phe Leu Ser Ser Ser Lys Gln Ile Val
            580                 585                 590

Ser Asp Met Asn Glu Lys Asp Lys Val Ile Val Phe Glu Arg Gly Asp
        595                 600                 605

Leu Val Phe Val Phe Asn Phe His Pro Asn Lys Thr Tyr Lys Gly Tyr
    610                 615                 620

Lys Val Gly Cys Asp Leu Pro Gly Lys Tyr Arg Val Ala Leu Asp Ser
625                 630                 635                 640

Asp Ala Leu Val Phe Gly Gly His Gly Arg Val Gly His Asp Val Asp
                645                 650                 655

His Phe Thr Ser Pro Glu Gly Met Pro Gly Val Pro Glu Thr Asn Phe
            660                 665                 670

Asn Asn Arg Pro Asn Ser Phe Lys Val Leu Ser Pro Pro Arg Thr Cys
        675                 680                 685

Val Ala Tyr Tyr Arg Val Asp Glu Asp Arg Glu Glu Leu Arg Arg Gly
    690                 695                 700

Gly Ala Val Ala Ser Gly Lys Ile Val Thr Glu Tyr Ile Asp Val Glu
705                 710                 715                 720

Ala Thr Ser Gly Glu Thr Ile Ser Gly Gly Trp Lys Gly Ser Glu Lys
                725                 730                 735

Asp Asp Cys Gly Lys Lys Gly Met Lys Phe Val Phe Arg Ser Ser Asp
            740                 745                 750
```

Glu Asp Cys Lys
755

<210> 25
<211> 762
<212> PRT
<213> Pisum sativum

<400> 25
Thr Met Pro Ser Val Glu Glu Asp Phe Glu Asn Ile Gly Ile Leu Asn
1 5 10 15

Val Asp Ser Ser Leu Glu Pro Phe Lys Asp His Phe Lys Tyr Arg Leu
20 25 30

Lys Arg Tyr Leu His Gln Lys Lys Leu Ile Glu Glu Tyr Glu Gly Gly
35 40 45

Leu Gln Glu Phe Ala Lys Gly Tyr Leu Lys Phe Gly Phe Asn Arg Glu
50 55 60

Glu Asp Gly Ile Ser Tyr Arg Glu Trp Ala Pro Ala Ala Gln Glu Ala
65 70 75 80

Gln Ile Ile Gly Asp Phe Asn Gly Trp Asn Gly Ser Asn Leu His Met
85 90 95

Glu Lys Asp Gln Phe Gly Val Trp Ser Ile Gln Ile Pro Asp Ala Asp
100 105 110

Gly Asn Pro Ala Ile Pro His Asn Ser Arg Val Lys Phe Arg Phe Lys
115 120 125

His Ser Asp Gly Val Trp Val Asp Arg Ile Pro Ala Trp Ile Lys Tyr
130 135 140

Ala Thr Val Asp Pro Thr Arg Phe Ala Ala Pro Tyr Asp Gly Val Tyr
145 150 155 160

Trp Asp Pro Pro Leu Ser Glu Arg Tyr Gln Phe Lys His Pro Arg Pro
165 170 175

Pro Lys Pro Lys Ala Pro Arg Ile Tyr Glu Ala His Val Gly Met Ser
180 185 190

Ser Ser Glu Pro Arg Ile Asn Ser Tyr Arg Glu Phe Ala Asp Asp Val
195 200 205

Leu Pro Arg Ile Arg Glu Asn Asn Tyr Asn Thr Val Gln Leu Met Ala
210 215 220

Val Met Glu His Ser Tyr Tyr Ala Ser Phe Trp Tyr His Val Thr Lys
225 230 235 240

Pro Phe Phe Ala Val Ser Ser Arg Ser Gly Ser Pro Glu Asp Leu Lys
245 250 255

Tyr Leu Ile Asp Lys Ala His Ser Leu Gly Leu Asn Val Leu Met Asp
260 265 270

Val Ile His Ser His Ala Ser Asn Asn Val Thr Asp Gly Leu Asn Gly
275 280 285

Phe Asp Val Gly Gln Ser Ser Gln Gln Ser Tyr Phe His Ala Gly Asp
290 295 300

Arg Gly Tyr His Lys Leu Trp Asp Ser Arg Leu Phe Asn Tyr Ala Asn
305 310 315 320

Trp Lys Ser Ser Phe Leu Leu Ser Asn Leu Arg Trp Trp Leu Glu Glu
325 330 335

Tyr Lys Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser Met Leu Tyr
340 345 350

His His His Gly Ile Asn Met Ala Phe Thr Gly Asp Tyr Asn Glu Tyr
355 360 365

Phe Ser Glu Glu Thr Asp Val Asp Ala Val Val Tyr Leu Met Leu Ala
370 375 380

Asn Ser Leu Val His Asp Ile Leu Pro Asp Ala Thr Asp Ile Ala Glu
385 390 395 400

Asp Val Ser Gly Met Pro Gly Leu Gly Arg Pro Val Ser Glu Val Gly
405 410 415

Ile Gly Phe Asp Tyr Arg Leu Ala Met Ala Ile Pro Asp Lys Trp Ile
420 425 430

Asp Tyr Leu Lys Asn Lys Lys Asp Ser Glu Trp Ser Met Lys Glu Ile
435 440 445

Ser Leu Asn Leu Thr Asn Arg Arg Tyr Thr Glu Lys Cys Val Ser Tyr
450 455 460

Ala Glu Ser His Asp Gln Ser Ile Val Gly Asp Lys Thr Ile Ala Phe
465 470 475 480

Leu Leu Met Asp Glu Glu Met Tyr Ser Ser Met Ser Cys Leu Thr Met
485 490 495

Leu Ser Pro Thr Ile Glu Arg Gly Ile Ser Leu His Lys Met Ile His
500 505 510

Phe Ile Thr Leu Ala Leu Gly Gly Glu Gly Tyr Leu Asn Phe Met Gly
515 520 525

Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Glu Gly Asn
530 535 540

Gly Trp Ser Tyr Glu Lys Cys Arg Leu Thr Gln Trp Asn Leu Val Asp
545 550 555 560

Thr Asn His Leu Arg Tyr Lys Phe Met Asn Ala Phe Asp Arg Ala Met
565 570 575

Asn Leu Leu Asp Asp Lys Phe Ser Ile Leu Ala Ser Thr Lys Gln Ile
580 585 590

Val Ser Ser Thr Asn Asn Glu Asp Lys Val Ile Val Phe Glu Arg Gly
595 600 605

Asp Leu Val Phe Val Phe Asn Phe His Pro Glu Asn Thr Tyr Glu Gly
610 615 620

Tyr Lys Val Gly Cys Asp Leu Pro Gly Lys Tyr Arg Val Ala Leu Asp
625 630 635 640

Ser Asp Ala Thr Glu Phe Gly Gly His Gly Arg Val Gly His Asp Ala
645 650 655

Asp Gln Phe Thr Ser Pro Glu Gly Ile Pro Gly Ile Pro Glu Thr Asn
660 665 670

Phe Asn Asn Arg Pro Asn Ser Phe Lys Val Leu Ser Pro Pro His Thr
675 680 685

Cys Val Val Tyr Tyr Arg Val Asp Glu Arg Gln Glu Glu Ser Asn Asn
690 695 700

Pro Asn Leu Gly Ser Val Glu Glu Thr Phe Ala Ala Ala Asp Thr Asp
705 710 715 720

Val Ala Arg Ile Pro Asp Val Ser Met Glu Ser Glu Asp Ser Asn Leu
725 730 735

Asp Arg Ile Glu Asp Asn Ser Glu Asp Ala Val Asp Ala Gly Ile Leu
740 745 750

Lys Val Glu Arg Glu Val Val Gly Asp Asn
755 760

<210> 26
<211> 984
<212> DNA
<213> Triticum aestivum

<400> 26
atatgtatga tttcatggct ctggatagac cttcaactcc tcgcattgat cgtggcatag 60
cattacataa aatgatcagg cttgtcacca tgggtttagg tggcgaaggc tatcttaact 120
tcatgggaaa tgagtttggg catcctgaat ggatagattt tccaagaggt ccgcaaactc 180
ttccaaccgg caaagttctc cctggaaata acaatagtta tgataaatgc cgccgtagat 240
ttgatcttgg agatgcagat tttcttagat atcgtggtat gcaagagtcc gaccaggcaa 300
tgcagcatct tgaggaaaaa tatgggttta tgacatctga gcaccagtat gtttcacgga 360
aacatgagga agataaggtg atcatcttcg aaagaggaga tttggtattc gttttcaact 420
tccaccggag caatagcttt tttgactacc gtgttgggtg ttccaggcct gggaagtaca 480
aggtggcctt agactccgac gatgcactct ttggtggatt cagcaggctt gatcatgatg 540
tcgactactt cacaaccgaa catccgcatg acaacaggcc gcgctctttc tcggtgtaca 600
ctccgagcag aactgcggtc gtgtatgccc ttacagagta agaaccagca gctgcttgtt 660
acaaggcaaa gagagaactc cagagagctc gtggatcgtg agcgaagcga cgggcaacgg 720
cgcgaggctg ctctaagcgc catgactggg aggggatcgt gcctcttccc cagatgccag 780
gaggagcaga tggataggta gcttgttggt gagcgctcga aagaaaatgg acgggcctgg 840
gtgtttgtcg tgctgcacta ccctcctcct atcttgcaca ttcccggttg tctttgtaca 900
tataactaat aattgcccgt gcgctcaacg tgaacatata aatattctaa taataggtta 960
tcccgtgaaa aaaaaaaaaa aaaa 984

<210> 27
<211> 977
<212> DNA
<213> Triticum aestivum

<400> 27
atatgtatga tttcatggct ctggatagac cttcaactcc tcgcattgat cgtggcatag 60
cattacataa aatgatcagg cttgtcacca tgggtttagg tggcgaaggc tatcttaact 120
tcatgggaaa tgagtttggg catcctgaat ggatagattt tccaagaggt ccgcaaactc 180
ttccaaccgg caaagttctc cctggaaata acaatagtta tgataaatgc cgccgtagat 240
ttgatcttgg agatgcagat tttcttagat atcgtggtat gcaagagttc gaccaggcaa 300
tgcagcatct tgaggaaaaa tatgggttta tgacatctga gcaccagtat gtttcacgga 360
aacatgagga agataaggtg atcatcttcg aaagaggaga tttggtattt gttttcaact 420

tccactggag caatagcttt tttgactacc gtgttgggtg ttccaagcct gggaagtaca 480
aggtggcctt agactccgac gatgcactct ttggtggatt cagcaggctt gatcatgatg 540
tcgactactt cacaaccgaa catccgcatg acaataggcc gcgctctttc ttggtgtaca 600
ctcctagcag aactgcggtc gtgtatgccc ttacagagta agaaccagca gcggcttgtt 660
acaaggcaaa gagagaactc cagggagctc gtggattgtg agcgaagcga cgggcaactg 720
cgtgaggctg ctctaagcgc catgactggg aggggatcgt gcctcttccc ctgatgccag 780
gaggatcaga tggataggta gcttgttggt gagcgctcga aagaaaatgg acgggcctgg 840
gtgtttgtcg tgctgcactt aaccctcctc ctatgttgca cattcccggg tgtttttgta 900
catataacta ataattgccc gtgcgcttca acatgaacat ataaatattc tatataaaaa 960
aaaaaaaaaa aaaaaaa 977

<210> 28
<211> 212
<212> PRT
<213> Triticum aestivum

<400> 28
Met Tyr Asp Phe Met Ala Leu Asp Arg Pro Ser Thr Pro Arg Ile Asp
1 5 10 15

Arg Gly Ile Ala Leu His Lys Met Ile Arg Leu Val Thr Met Gly Leu
20 25 30

Gly Gly Glu Gly Tyr Leu Asn Phe Met Gly Asn Glu Phe Gly His Pro
35 40 45

Glu Trp Ile Asp Phe Pro Arg Gly Pro Gln Thr Leu Pro Thr Gly Lys
50 55 60

Val Leu Pro Gly Asn Asn Asn Ser Tyr Asp Lys Cys Arg Arg Arg Phe
65 70 75 80

Asp Leu Gly Asp Ala Asp Phe Leu Arg Tyr Arg Gly Met Gln Glu Phe
85 90 95

Asp Gln Ala Met Gln His Leu Glu Glu Lys Tyr Gly Phe Met Thr Ser
100 105 110

Glu His Gln Tyr Val Ser Arg Lys His Glu Glu Asp Lys Val Ile Ile
115 120 125

Phe Glu Arg Gly Asp Leu Val Phe Val Phe Asn Phe His Trp Ser Asn
130 135 140

Ser Phe Phe Asp Tyr Arg Val Gly Cys Ser Lys Pro Gly Lys Tyr Lys
145 150 155 160

Val Ala Leu Asp Ser Asp Asp Ala Leu Phe Gly Gly Phe Ser Arg Leu

165 170 175

Asp His Asp Val Asp Tyr Phe Thr Thr Glu His Pro His Asp Asn Arg
180 185 190

Pro Arg Ser Phe Leu Val Tyr Thr Pro Ser Arg Thr Ala Val Val Tyr
195 200 205

Ala Leu Thr Glu
210

<210> 29
<211> 212
<212> PRT
<213> Zea mays

<400> 29
Met Tyr Asp Phe Met Ala Leu Asp Arg Pro Ser Thr Pro Thr Ile Asp
1 5 10 15

Arg Gly Ile Ala Leu His Lys Met Ile Arg Leu Ile Thr Met Gly Leu
20 25 30

Gly Gly Glu Gly Tyr Leu Asn Phe Met Gly Asn Glu Phe Gly His Pro
35 40 45

Glu Trp Ile Asp Phe Pro Arg Gly Pro Gln Arg Leu Pro Ser Gly Lys
50 55 60

Phe Ile Pro Gly Asn Asn Asn Ser Tyr Asp Lys Cys Arg Arg Arg Phe
65 70 75 80

Asp Leu Gly Asp Ala Asp Tyr Leu Arg Tyr His Gly Met Gln Glu Phe
85 90 95

Asp Gln Ala Met Gln His Leu Glu Gln Lys Tyr Glu Phe Met Thr Ser
100 105 110

Asp His Gln Tyr Ile Ser Arg Lys His Glu Glu Asp Lys Val Ile Val
115 120 125

Phe Glu Lys Gly Asp Leu Val Phe Val Phe Asn Phe His Cys Asn Asn
130 135 140

Ser Tyr Phe Asp Tyr Arg Ile Gly Cys Arg Lys Pro Gly Val Tyr Lys
145 150 155 160

Val Val Leu Asp Ser Asp Ala Gly Leu Phe Gly Gly Phe Ser Arg Ile
165 170 175

His His Ala Ala Glu His Phe Thr Ala Asp Cys Ser His Asp Asn Arg
180 185 190

Pro Tyr Ser Phe Ser Val Tyr Thr Pro Ser Arg Thr Cys Val Val Tyr
195 200 205

Ala Pro Val Glu
210

<210> 30
<211> 216
<212> PRT
<213> Zea mays

<400> 30
Met Tyr Asp Phe Met Ala Leu Asp Arg Pro Ser Thr Pro Arg Ile Asp
1 5 10 15

Arg Gly Ile Ala Leu His Lys Met Ile Arg Leu Val Thr Met Gly Leu
20 25 30

Gly Gly Glu Gly Tyr Leu Asn Phe Met Gly Asn Glu Phe Gly His Pro
35 40 45

Glu Trp Ile Asp Phe Pro Arg Gly Pro Gln Ser Leu Pro Asn Gly Ser
50 55 60

Val Ile Pro Gly Asn Asn Asn Ser Phe Asp Lys Cys Arg Arg Arg Phe
65 70 75 80

Asp Leu Gly Asp Ala Asp Tyr Leu Arg Tyr Arg Gly Met Gln Glu Phe
85 90 95

Asp Gln Ala Met Gln His Leu Glu Gly Lys Tyr Glu Phe Met Thr Ser
100 105 110

Asp His Ser Tyr Phe Ser Arg Lys His Glu Glu Asp Lys Val Ile Ile
115 120 125

Phe Glu Arg Gly Asp Leu Val Phe Val Phe Asn Phe His Trp Ser Asn
130 135 140

Ser Tyr Phe Asp Tyr Arg Val Gly Cys Phe Lys Pro Gly Lys Tyr Lys
145 150 155 160

Ile Val Leu Asp Ser Asp Asp Gly Leu Phe Gly Gly Phe Ser Arg Leu
165 170 175

Asp His Asp Ala Glu Tyr Phe Thr Ala Asp Trp Pro His Asp Asn Arg
180 185 190

Pro Cys Ser Phe Ser Val Tyr Ala Pro Ser Arg Thr Ala Val Val Tyr
195 200 205

Ala Pro Ala Gly Ala Glu Asp Glu
210 215

<210> 31
<211> 217
<212> DNA
<213> Zea mays

<400> 31
tagcggggta ctcgttgctg cgcggcatgt gtggggctgt cgatgtgagg aaaaaccttc 60
ttccaaaacc ggcagatgca tgcatgcatg ctacaataag gttctgatac tttaatcgat 120
gctggaaagc ccatgcatct cgctgcgttg tcctctctat atatataaga ccttcaaggt 180
gtcaattaaa catagagttt tcgtttttcg ctttcct 217

<210> 32
<211> 686
<212> PRT
<213> Triticum aestivum

<400> 32
Met Leu Cys Leu Ser Ser Ser Leu Leu Pro Arg Pro Ser Ala Ala Ala
1 5 10 15

Asp Arg Pro Leu Pro Gly Ile Ile Ala Gly Gly Gly Gly Gly Lys Arg
20 25 30

Leu Ser Val Val Pro Ser Val Pro Phe Leu Leu Arg Trp Leu Trp Pro
35 40 45

Arg Lys Ala Lys Ser Lys Ser Phe Val Ser Val Thr Ala Arg Gly Asn
50 55 60

Lys Ile Ala Ala Thr Thr Gly Tyr Gly Ser Asp His Leu Pro Ile Tyr
65 70 75 80

Asp Leu Asp Leu Lys Leu Ala Glu Phe Lys Asp His Phe Asp Tyr Thr
85 90 95

Arg Asn Arg Tyr Ile Glu Gln Lys His Leu Ile Glu Lys His Glu Gly
100 105 110

Ser Leu Glu Glu Phe Ser Lys Gly Tyr Leu Lys Phe Gly Ile Asn Thr
115 120 125

Glu His Gly Ala Ser Val Tyr Arg Glu Trp Ala Pro Ala Ala Glu Glu
130 135 140

Ala Gln Leu Val Gly Asp Phe Asn Asn Trp Asn Gly Ser Gly His Lys
145 150 155 160

Met Ala Lys Asp Asn Phe Gly Val Trp Ser Ile Arg Ile Ser His Val
165 170 175

Asn Gly Lys Pro Ala Ile Pro His Asn Ser Lys Val Lys Phe Arg Phe
180 185 190

Arg His His Gly Val Trp Val Glu Gln Ile Pro Ala Trp Ile Arg Tyr
195 200 205

Ala Thr Val Thr Ala Ser Glu Ser Gly Ala Pro Tyr Asp Gly Leu His
210 215 220

Trp Asp Pro Pro Ser Ser Glu Arg Tyr Val Phe Asn His Pro Arg Pro
225 230 235 240

Pro Lys Pro Asp Val Pro Arg Ile Tyr Glu Ala His Val Gly Val Ser
245 250 255

Gly Gly Lys Leu Glu Ala Gly Thr Tyr Arg Glu Phe Pro Asp Asn Val
260 265 270

Leu Pro Cys Leu Arg Ala Thr Asn Tyr Asn Thr Val Gln Leu Met Gly
275 280 285

Ile Met Glu His Ser Asp Ser Ala Ser Phe Gly Tyr His Val Thr Asn
290 295 300

Phe Phe Ala Val Ser Ser Arg Ser Gly Thr Pro Glu Asp Leu Lys Tyr
305 310 315 320

Leu Ile Asp Lys Ala His Ser Leu Gly Leu Arg Val Leu Met Asp Val
325 330 335

Val His Ser His Ala Ser Asn Asn Val Ile Asp Gly Leu Asn Gly Tyr
340 345 350

Asp Val Gly Gln Ser Ala His Glu Ser Tyr Phe Tyr Thr Gly Asp Lys
355 360 365

Gly Tyr Asn Lys Met Trp Asn Gly Arg Met Phe Asn Tyr Ala Asn Trp
370 375 380

Glu Val Leu Arg Phe Leu Leu Ser Asn Leu Arg Tyr Trp Met Asp Glu
385 390 395 400

Phe Met Phe Asp Gly Phe Arg Phe Val Gly Val Thr Ser Met Leu Tyr
405 410 415

Asn His Asn Gly Ile Asn Met Ser Phe Asn Gly Asn Tyr Lys Asp Tyr
420 425 430

Ile Gly Leu Asp Thr Asn Val Asp Ala Phe Val Tyr Met Met Leu Ala
435 440 445

Asn His Leu Met His Lys Leu Phe Pro Glu Ala Ile Val Val Ala Val
450 455 460

Asp Val Ser Gly Met Pro Val Leu Cys Trp Pro Val Asp Glu Gly Gly
465 470 475 480

Leu Gly Phe Asp Tyr Arg Gln Ala Met Thr Ile Pro Asp Arg Trp Ile
485 490 495

Asp Tyr Leu Glu Asn Lys Gly Asp Gln Gln Trp Ser Met Ser Ser Val
500 505 510

Ile Ser Gln Thr Leu Thr Asn Arg Arg Tyr Pro Glu Lys Phe Ile Ala
515 520 525

Tyr Ala Glu Arg Gln Asn His Ser Ile Ile Gly Ser Lys Thr Met Ala
530 535 540

Phe Leu Leu Met Glu Trp Glu Thr Tyr Ser Gly Met Ser Ala Met Asp
545 550 555 560

Pro Asp Ser Pro Thr Ile Asp Arg Ala Ile Ala Leu Gln Lys Met Ile
565 570 575

His Phe Ile Thr Met Ala Phe Gly Gly Asp Ser Tyr Leu Lys Phe Met
580 585 590

Gly Asn Glu Tyr Met Asn Ala Phe Val Gln Ala Val Asp Thr Pro Ser
595 600 605

Asp Lys Cys Ser Phe Leu Ser Ser Ser Asn Gln Thr Ala Ser His Met
610 615 620

Asn Glu Glu Glu Lys Gly Ser Ala Leu Thr Lys Gly Tyr Thr His Leu
625 630 635 640

Arg Ser Gly Cys Phe Asp Pro Ser Leu Pro Ser Thr Ser Ser Cys Ala
645 650 655

Phe Leu Gly Pro Ser Asn Gln Ser Pro Phe Ser Lys Pro Phe Ile Gly
660 665 670

Phe Pro Gly Cys Ile Phe Cys Cys Gly Leu Phe Lys Gly Glu
675 680 685

<210> 33
<211> 830
<212> PRT
<213> Triticum aestivum

<400> 33
Met Leu Cys Leu Thr Ala Pro Ser Cys Ser Pro Ser Leu Pro Pro Arg
1 5 10 15

Pro Ser Arg Pro Ala Ala Asp Arg Pro Gly Pro Gly Ile Ser Gly Gly
20 25 30

Gly Asn Val Arg Leu Ser Ala Val Pro Ala Pro Ser Ser Leu Arg Trp
35 40 45

Ser Trp Pro Arg Lys Ala Lys Ser Lys Phe Ser Val Pro Val Ser Ala
50 55 60

Pro Arg Asp Tyr Thr Met Ala Thr Ala Glu Asp Gly Val Gly Asp Leu
65 70 75 80

Pro Ile Tyr Asp Leu Asp Pro Lys Phe Ala Gly Phe Lys Glu His Phe
85 90 95

Ser Tyr Arg Met Lys Lys Tyr Leu Asp Gln Lys His Ser Ile Glu Lys
100 105 110

His Glu Gly Gly Leu Glu Glu Phe Ser Lys Gly Tyr Leu Lys Phe Gly
115 120 125

Ile Asn Thr Glu Asn Asp Ala Thr Val Tyr Arg Glu Trp Ala Pro Ala

130 135 140

Ala Met Asp Ala Gln Leu Ile Gly Asp Phe Asn Asn Trp Asn Gly Ser
145 150 155 160

Gly His Arg Met Thr Lys Asp Asn Tyr Gly Val Trp Ser Ile Arg Ile
165 170 175

Ser His Val Asn Gly Lys Pro Ala Ile Pro His Asn Ser Lys Val Lys
180 185 190

Phe Arg Phe His Arg Gly Asp Gly Leu Trp Val Asp Arg Val Pro Ala
195 200 205

Trp Ile Arg Tyr Ala Thr Phe Asp Ala Ser Lys Phe Gly Ala Pro Tyr
210 215 220

Asp Gly Val His Trp Asp Pro Pro Ser Gly Glu Arg Tyr Val Phe Lys
225 230 235 240

His Pro Arg Pro Arg Lys Pro Asp Ala Pro Arg Ile Tyr Glu Ala His
245 250 255

Val Gly Met Ser Gly Glu Lys Pro Glu Val Ser Thr Tyr Arg Glu Phe
260 265 270

Ala Asp Asn Val Leu Pro Arg Ile Lys Ala Asn Asn Tyr Asn Thr Val
275 280 285

Gln Leu Met Ala Ile Met Glu His Ser Tyr Tyr Ala Ser Phe Gly Tyr
290 295 300

His Val Thr Asn Phe Phe Ala Val Ser Ser Arg Ser Gly Thr Pro Glu
305 310 315 320

Asp Leu Lys Tyr Leu Val Asp Lys Ala His Ser Leu Gly Leu Arg Val
325 330 335

Leu Met Asp Val Val His Ser His Ala Ser Ser Asn Lys Thr Asp Gly
340 345 350

Leu Asn Gly Tyr Asp Val Gly Gln Asn Thr Gln Glu Ser Tyr Phe His
355 360 365

Thr Gly Glu Arg Gly Tyr His Lys Leu Trp Asp Ser Arg Leu Phe Asn
370 375 380

Tyr Ala Asn Trp Glu Val Leu Arg Phe Leu Leu Ser Asn Leu Arg Tyr

385 390 395 400

Trp Met Asp Glu Phe Met Phe Asp Gly Phe Arg Phe Asp Gly Val Thr
405 410 415

Ser Met Leu Tyr Asn His His Gly Ile Asn Met Ser Phe Ala Gly Ser
420 425 430

Tyr Lys Glu Tyr Phe Gly Leu Asp Thr Asp Val Asp Ala Val Val Tyr
435 440 445

Leu Met Leu Ala Asn His Leu Met His Lys Leu Leu Pro Glu Ala Thr
450 455 460

Val Val Ala Glu Asp Val Ser Gly Met Pro Val Leu Cys Arg Ser Val
465 470 475 480

Asp Glu Gly Gly Val Gly Phe Asp Tyr Arg Leu Ala Met Ala Ile Pro
485 490 495

Asp Arg Trp Ile Asp Tyr Leu Lys Asn Lys Asp Asp Leu Glu Trp Ser
500 505 510

Met Ser Gly Ile Ala His Thr Leu Thr Asn Arg Arg Tyr Thr Glu Lys
515 520 525

Cys Ile Ala Tyr Ala Glu Ser His Asp Gln Ser Ile Val Gly Asp Lys
530 535 540

Thr Met Ala Phe Leu Leu Met Asp Lys Glu Met Tyr Thr Gly Met Ser
545 550 555 560

Asp Leu Gln Pro Ala Ser Pro Thr Ile Asp Arg Gly Ile Ala Leu Gln
565 570 575

Lys Met Ile His Phe Ile Thr Met Ala Leu Gly Gly Asp Gly Tyr Leu
580 585 590

Asn Phe Met Gly Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro
595 600 605

Arg Glu Gly Asn Asn Trp Ser Tyr Asp Lys Cys Arg Arg Gln Trp Ser
610 615 620

Leu Ala Asp Ile Asp His Leu Arg Tyr Lys Tyr Met Asn Ala Phe Asp
625 630 635 640

Gln Ala Met Asn Ala Leu Asp Asp Lys Phe Ser Phe Leu Ser Ser Ser

645 650 655

Lys Gln Ile Val Ser Asp Met Asn Glu Glu Lys Lys Ile Ile Val Phe
660 665 670

Glu Arg Gly Asp Leu Val Phe Val Phe Asn Phe His Pro Ser Lys Thr
675 680 685

Tyr Asp Gly Tyr Lys Val Gly Cys Asp Leu Pro Gly Lys Tyr Lys Val
690 695 700

Ala Leu Asp Ser Asp Ala Leu Met Phe Gly Gly His Gly Arg Val Ala
705 710 715 720

His Asp Asn Asp His Phe Thr Ser Pro Glu Gly Val Pro Gly Val Pro
725 730 735

Glu Thr Asn Phe Asn Asn Arg Pro Asn Ser Phe Lys Ile Leu Ser Pro
740 745 750

Ser Arg Thr Cys Val Ala Tyr Tyr Arg Val Glu Glu Lys Ala Glu Lys
755 760 765

Pro Lys Asp Glu Gly Ala Ala Ser Trp Gly Lys Thr Ala Leu Gly Tyr
770 775 780

Ile Asp Val Glu Ala Thr Gly Val Lys Asp Ala Ala Asp Gly Glu Ala
785 790 795 800

Thr Ser Gly Ser Glu Lys Ala Ser Thr Gly Gly Asp Ser Ser Lys Lys
805 810 815

Gly Ile Asn Phe Val Phe Leu Ser Pro Asp Lys Asp Asn Lys
820 825 830

<210> 34
<211> 818
<212> PRT
<213> Triticum aestivum

<400> 34
Met Ala Thr Phe Ala Val Ser Gly Trp Thr Leu Gly Val Ala Arg Pro
1 5 10 15

Ala Gly Ala Gly Gly Gly Leu Leu Pro Arg Ser Gly Ser Glu Arg Arg
20 25 30

Gly Gly Val Asp Leu Pro Ser Leu Leu Leu Arg Lys Lys Asp Ser Ser
35 40 45

Arg Ala Ala Ser Pro Gly Lys Val Leu Val Pro Asp Gly Glu Ser Asp
50 55 60

Asp Leu Ala Ser Pro Ala Gln Pro Glu Glu Leu Gln Ile Pro Glu Asp
65 70 75 80

Ile Glu Glu Gln Thr Ala Glu Val Asn Met Thr Gly Gly Thr Ala Glu
85 90 95

Lys Leu Glu Ser Ser Glu Pro Thr Gln Gly Ile Val Glu Thr Ile Thr
100 105 110

Asp Gly Val Thr Lys Gly Val Lys Glu Leu Val Val Gly Glu Lys Pro
115 120 125

Arg Val Val Pro Lys Pro Gly Asp Gly Gln Lys Ile Tyr Glu Ile Asp
130 135 140

Pro Thr Leu Lys Asp Phe Arg Ser His Leu Asp Tyr Arg Tyr Ser Glu
145 150 155 160

Tyr Arg Arg Ile Arg Ala Ala Ile Asp Gln His Glu Gly Gly Leu Glu
165 170 175

Ala Phe Ser Arg Gly Tyr Glu Lys Leu Gly Phe Thr Arg Ser Ala Glu
180 185 190

Gly Ile Thr Tyr Arg Glu Trp Ala Pro Gly Ala His Ser Ala Ala Leu
195 200 205

Val Gly Asp Phe Asn Asn Trp Asn Pro Asn Ala Asp Thr Met Thr Arg
210 215 220

Asp Asp Tyr Gly Val Trp Glu Ile Phe Leu Pro Asn Asn Ala Asp Gly
225 230 235 240

Ser Pro Ala Ile Pro His Gly Ser Arg Val Lys Ile Arg Met Asp Thr
245 250 255

Pro Ser Gly Val Lys Asp Ser Ile Ser Ala Trp Ile Lys Phe Ser Val
260 265 270

Gln Ala Pro Gly Glu Ile Pro Phe Asn Gly Ile Tyr Tyr Asp Pro Pro
275 280 285

Glu Glu Glu Lys Tyr Val Phe Gln His Pro Gln Pro Lys Arg Pro Glu
290 295 300

Ser Leu Arg Ile Tyr Glu Ser His Ile Gly Met Ser Ser Pro Glu Pro
305 310 315 320

Lys Ile Asn Ser Tyr Ala Asn Phe Arg Asp Glu Val Leu Pro Arg Ile
325 330 335

Lys Arg Leu Gly Tyr Asn Ala Val Gln Ile Met Ala Ile Gln Glu His
340 345 350

Ser Tyr Tyr Ala Ser Phe Gly Tyr His Val Thr Asn Phe Phe Ala Pro
355 360 365

Ser Ser Arg Phe Gly Thr Pro Glu Asp Leu Lys Ser Leu Ile Asp Arg
370 375 380

Ala His Glu Leu Gly Leu Ile Val Leu Met Asp Ile Val His Ser His
385 390 395 400

Ser Ser Asn Asn Thr Leu Asp Gly Leu Asn Gly Phe Asp Gly Thr Asp
405 410 415

Thr His Tyr Phe His Gly Gly Pro Arg Gly His His Trp Met Trp Asp
420 425 430

Ser Arg Leu Phe Asn Tyr Gly Ser Trp Glu Val Leu Arg Phe Leu Leu
435 440 445

Ser Asn Ala Arg Trp Trp Leu Glu Glu Tyr Lys Phe Asp Gly Phe Arg
450 455 460

Phe Asp Gly Val Thr Ser Met Met Tyr Thr His His Gly Leu Gln Met
465 470 475 480

Thr Phe Thr Gly Asn Tyr Gly Glu Tyr Phe Gly Phe Ala Thr Asp Val
485 490 495

Asp Ala Val Val Tyr Leu Met Leu Val Asn Asp Leu Ile His Gly Leu
500 505 510

His Pro Asp Ala Val Ser Ile Gly Glu Asp Val Ser Gly Met Pro Thr
515 520 525

Phe Cys Ile Pro Val Pro Asp Gly Gly Val Gly Leu Asp Tyr Arg Leu
530 535 540

His Met Ala Val Ala Asp Lys Trp Ile Glu Leu Leu Lys Gln Ser Asp
545 550 555 560

Glu Ser Trp Lys Met Gly Asp Ile Val His Thr Leu Thr Asn Arg Arg
565 570 575

Trp Leu Glu Lys Cys Val Thr Tyr Ala Glu Ser His Asp Gln Ala Leu
580 585 590

Val Gly Asp Lys Thr Ile Ala Phe Trp Leu Met Asp Lys Asp Met Tyr
595 600 605

Asp Phe Met Ala Leu Asp Arg Pro Ser Thr Pro Arg Ile Asp Arg Gly
610 615 620

Ile Ala Leu His Lys Met Ile Arg Leu Val Thr Met Gly Leu Gly Gly
625 630 635 640

Glu Gly Tyr Leu Asn Phe Met Gly Asn Glu Phe Gly His Pro Glu Trp
645 650 655

Ile Asp Phe Pro Arg Gly Pro Gln Thr Leu Pro Thr Gly Lys Val Leu
660 665 670

Pro Gly Asn Asn Asn Ser Tyr Asp Lys Cys Arg Arg Arg Phe Asp Leu
675 680 685

Gly Asp Ala Asp Phe Leu Arg Tyr His Gly Met Gln Glu Phe Asp Gln
690 695 700

Ala Met Gln His Leu Glu Glu Lys Tyr Gly Phe Met Thr Ser Glu His
705 710 715 720

Gln Tyr Val Ser Arg Lys His Glu Glu Asp Lys Val Ile Ile Phe Glu
725 730 735

Arg Gly Asp Leu Val Phe Val Phe Asn Phe His Trp Ser Asn Ser Phe
740 745 750

Phe Asp Tyr Arg Val Gly Cys Ser Arg Pro Gly Lys Tyr Lys Val Ala
755 760 765

Leu Asp Ser Asp Asp Ala Leu Phe Gly Gly Phe Ser Arg Leu Asp His
770 775 780

Asp Val Asp Tyr Phe Thr Thr Glu His Pro His Asp Asn Arg Pro Arg
785 790 795 800

Ser Phe Ser Val Tyr Thr Pro Ser Arg Thr Ala Val Val Tyr Ala Leu
805 810 815

Thr Glu

<210> 35
<211> 813
<212> DNA
<213> Escherichia coli

<400> 35
gagctccgtt tcgcatgatt gaacaagatg gattgcacgc aggttctccg gccgcttggg 60
tggagaggct attcggctat gactgggcac aacagacaat cggctgctct gatgccgccg 120
tgttccggct gtcagcgcag gggcgcccgg ttctttttgt caagaccgac ctgtccggtg 180
ccctgaatga actgcaggac gaggcagcgc ggctatcgtg gctggccacg acgggcgttc 240
cttgcgcagc tgtgctcgac gttgtcactg aagcgggaag ggactggctg ctattgggcg 300
aagtgccggg gcaggatctc ctgtcatctc accttgctcc tgccgagaaa gtatccatca 360
tggctgatgc aatgcggcgg ctgcatacgc ttgatccggc tacctgccca ttcgaccacc 420
aagcgaaaca tcgcatcgag cgagcacgta ctcggatgga agccggtctt gtcgatcagg 480
atgatctgga cgaagagcat caggggctcg cgccagccga actgttcgcc aggctcaagg 540
cgcgcatgcc cgacggcgag gatctcgtcg tgacccatgg cgatgcctgc ttgccgaata 600
tcatggtgga aaatggccgc ttttctggat tcatcgactg tggccggctg ggtgtggcgg 660
accgctatca ggacatagcg ttggctaccc gtgatattgc tgaagagctt ggcggcgaat 720
gggctgaccg cttcctcgtg ctttacggta tcgccgctcc cgattcgcag cgcatcgcct 780
tctatcgcct tcttgacgag ttcttctgag ctc 813

<210> 36
<211> 7
<212> PRT
<213> Triticum aestivum

<400> 36
Met Asp Lys Asp Met Tyr Asp
1 5

<210> 37
<211> 40
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 37

aaggatccgt cgacatcgat aatacgactc actataggga 40

<210> 38
<211> 17
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 38
aaggatccgt cgacatc 17

<210> 39
<211> 20
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 39
atggacaagg atatgtatga 20

<210> 40
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 40
ttttcttcac aacgccctgg g 21

<210> 41
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 41
tgtttgggag atcttcctcc c 21

<210> 42
<211> 8
<212> PRT
<213> Triticum aestivum

<400> 42
Gly Val Trp Glu Ile Phe Leu Pro
1 5

<210> 43
<211> 10
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 43
cgggatcccg 10

<210> 44
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 44
gatgagctcc gtttcgcatg attgaacaag atgg 34

<210> 45
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 45
gtcgagctca gaagaactcg tcaagaaggc 30

<210> 46
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 46
cccgacggcg aggatctcgt gctgacc 27

<210> 47
<211> 35
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 47
catgggtcac gacgagatcc tcgccgtcgg gcatg 35

<210> 48
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 48
attaggtacc ggacttgctc cgctgtcggc 30

<210> 49
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 49
tataggtacc gaggcagcga cagagatgcc 30

<210> 50
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 50
agctgaatcc ggcggcatgg c 21

<210> 51
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:synthetic
oligonucleotide

<400> 51
tgatagtctt gccagtcagg g 21

<210> 52
<211> 2037
<212> DNA
<213> Zea mays

<400> 52
ttagctgaat ccggcggcat ggcaaggtag actgcagtgc agcgtgaccc ggtcgtgccc 60
ctctctagag ataatgagca ttgcatgtct aagttataaa aaattaccac atattttttt 120
tgtcacactt gtttgaagtg cagtttatct atctttatac atatatttaa actttactct 180
acgaataata taatctatag tactacaata atatcagtgt tttagagaat catataaatg 240
aacagttaga catggtctaa aggacaattg gtattttgac aacaggactc tacagtttta 300
tctttttagt gtgcatgtgt tctccttttt ttttttgcaa atagcttcac ctatataata 360
cttcatccat tttattagta catccattta gggtttaggg ttaatggttt ttatagacta 420
attttttag tacatctatt ttattctatt ttagcctcta aattaagaaa actaaaactc 480
tattttagtt tttttattta ataatttaga tataaaatag aataaaataa agtgactaaa 540
aattaaacaa ataccctta agaaattaaa aaaactaagg aaacattttt cttgtttcga 600
gtagataatg ccagcctgtt aaacgccgtc gacgcagtct aacggacacc aaccagcgaa 660
ccagcagcgt cgcgtcgggc caagcgaagc agacggcacg gcatctctgt cgctgcctcg 720
gtaccggact tcgtccgctg tcggcatcca gaaattgcgt ggcggagcgg cagacgtgag 780
ccggcacggc aggcggcctc ctcctcctct cacggcaccg gcagctacgg gggattcctt 840
tcccaccgct ccttcgcttt cccttcctcg cccgccgtaa taaatagaca ccccctccac 900
accctctttc cccaacctcg tgttgttcgg agcgcacaca cacacaacca gatctccccc 960
aaatccaccc gtcggcacct ccgcttcaag gtacgccgct cgtcctcccc ccccctctct 1020

acctctcta gatcggcgtt ccggtccatg gttagggccc ggtagttcta cttctgttca 1080
tgtttgtgtt agatccgtgt ttgtgttaga tccgtgctgc tagcgttcgt acacggatgc 1140
gacctgtacg tcagacacgt tctgattgct aacttgccag tgtttctctt tggggaatcc 1200
tgggatggct ctagccgttc cgcagacggg atcgatttca tgattttttt tgtttcgttg 1260
catagggttt ggtttgccct tttcctttat ttcaatatat gccgtgcact tgtttgtcgg 1320
gtcatctttt catgcttttt tttgtcttgg ttgtgatgat gtggtctggt tgggcggtcg 1380
ttctagatcg gagtagaatt ctgtttcaaa ctacctggtg gatttattaa ttttggatct 1440
gtatgtgtgt gccatacata ttcatagtta cgaattgaag atgatggatg gaaatatcga 1500
tctaggatag gtatacatgt tgatgcgggt tttactgatg catatacaga gatgcttttg 1560
ttcgcttggt tgtgatgatg tggtgtggtt gggcggtcgt tcattcgttc tagatcggag 1620
tagaatactg tttcaaacta cctggtgtat ttattaattt tggaactgta tgtgtgtgtc 1680
atacatcttc atagttacga gtttaagatg gatggaaata tcgatctagg ataggtatac 1740
atgttgatgt gggttttact gatgcatata catgatggca tatgcagcat ctattcatat 1800
gctctaacct tgagtaccta tctattataa taaacaagta tgttttataa ttattttgat 1860
cttgatatac ttggatgatg gcatatgcag cagctatatg tggatttttt tagccctgcc 1920
ttcatacgct atttatttgc ttggtactgt ttcttttgtc gatgctcacc ctgttgtttg 1980
gtgttacttc tgcagatgca gatctttgtg aaaaccctga ctggcaagac tatcacc 2037

<210> 53
<211> 1085
<212> DNA
<213> Triticum aestivum

<400> 53
atatgtatga tttcatggct ctggatggac cttcgactcc tgctattgat cgtggcatag 60
cattgcataa aatgattagg cttgtcacca tgggtttagg tggagagggt tatcttaact 120
ttatgggaaa tgagtttggg catcctgaat ggatagattt tccaagaggc ccacaagttc 180
ttccaactgg taagtttctc cctggaaata acaatagtta tgataaatgc cgtcgtagat 240
ttgatcttgg tgatgcagat ttttcttaggt atcgtggtat gcaggagttt gatcaggcaa 300
tgcagcatct tgaggaaaaa tatgggttta tgacatctga gcaccagtat gtttctcgga 360
aacatgagga agataaggtg atcgtgtttg aaagagggga tttggtattt gttttcaact 420
tccactggag taatagcttt tttgactacc gtgttgggtg tttcaagcct gggaagtaca 480
aggtggtctt agactccgac gctggactct ttggtggatt tggtaggctt gatcatgctg 540
tcgagtactt cacttctgac tgtccgcatg acaacaggcc gcattctttc tcggtgtaca 600
ctcctagcag aacttgtgtt gtgtatgctc ttatggagta agcagcaagt gcagcatacg 660
ctgccgctgt tgttgctagt agcaaggaga gatcgtaggt cactacacca ggtgcagggt 720
ttgatatgga tttttgcttg agcgagtcct ggatgggcaa gacagcgtga tgctgtgtgt 780
gctcccaaat cgccatggcg ttgggagggg atcgtgcttc tttgtgttat gctttgtgga 840
tcaggatgga actcccctag gtagccttgt tggtgagcgc tcgaaagaaa atggacgggc 900
ctgggtgttt gcttaaattt tgttgcccta aaccctcgct cctatcttgt acattgccgg 960
tttagatagg gtttgttttt gtacattttt ttgatagtta atagacttat tgctcgtgtg 1020
cttgacgttt tacatgaaca tataaatatt ctaaataggt taaaaaaaaa aaaaaaaaaa 1080
aaaaa 1085

<210> 54
<211> 888
<212> PRT
<213> Triticum aestivum

<400> 54

Met Leu Cys Leu Ser Xaa Ser Leu Leu Pro Arg Pro Ser Arg Ala Ala
1 5 10 15

Ala Asp Arg Pro Xaa Leu Pro Gly Ile Xaa Gly Gly Gly Xaa Xaa Arg
20 25 30

Leu Ser Ala Val Pro Ala Pro Xaa Xaa Leu Arg Trp Xaa Trp Pro Arg
35 40 45

Lys Ala Lys Ser Lys Ser Ser Val Pro Val Xaa Ala Xaa Xaa Xaa Xaa
50 55 60

Ile Xaa Ala Thr Xaa Xaa Xaa Gly Val Xaa Xaa Leu Pro Ile Tyr Asp
65 70 75 80

Leu Asp Pro Lys Leu Ala Xaa Phe Lys Xaa His Phe Asp Tyr Arg Xaa
85 90 95

Xaa Xaa Tyr Xaa Xaa Gln Lys His Xaa Ile Glu Lys His Glu Gly Gly
100 105 110

Leu Glu Glu Phe Ser Lys Gly Tyr Leu Lys Phe Gly Ile Asn Thr Glu
115 120 125

Xaa Xaa Ala Xaa Val Tyr Arg Glu Trp Ala Pro Ala Ala Xaa Xaa Ala
130 135 140

Gln Leu Val Gly Asp Phe Asn Asn Trp Asn Gly Ser Gly His Xaa Met
145 150 155 160

Thr Lys Asp Asn Phe Gly Val Trp Ser Ile Arg Leu Ser Asn Asn Ala
165 170 175

Asp Gly Ser Pro Ala Ile Pro His Gly Ser Lys Val Lys Phe Arg Phe
180 185 190

Asp Thr Pro Ser Gly Val Trp Val Asp Ser Ile Pro Ala Trp Ile Lys
195 200 205

Tyr Ala Val Gln Thr Ala Gly Glu Ile Gly Ala Pro Tyr Asp Gly Ile
210 215 220

His Tyr Asp Pro Pro Ser Glu Glu Lys Tyr Val Phe Lys His Pro Gln
225 230 235 240

Pro Lys Lys Pro Asp Ser Leu Arg Ile Tyr Glu Ala His Val Gly Met

245 250 255

Ser Gly Pro Glu Pro Glu Ile Asn Thr Tyr Ala Glu Phe Arg Asp Glu
260 265 270

Val Leu Pro Arg Ile Lys Ala Leu Gly Tyr Asn Ala Val Gln Leu Met
275 280 285

Ala Ile Gln Glu His Ser Tyr Tyr Ala Ser Phe Gly Tyr His Val Thr
290 295 300

Asn Phe Phe Ala Val Ser Ser Arg Ser Gly Thr Pro Glu Asp Leu Lys
305 310 315 320

Ser Leu Ile Asp Lys Ala His Ser Leu Gly Leu Arg Val Leu Met Asp
325 330 335

Val Val His Ser His Ala Ser Asn Asn Thr Leu Asp Gly Leu Asn Gly
340 345 350

Phe Asp Val Gly Gln Gly Thr Asp Thr Ser Tyr Phe His Gly Gly Xaa
355 360 365

Arg Gly His His Lys Met Trp Asp Ser Arg Leu Phe Asn Tyr Gly Asn
370 375 380

Trp Glu Val Leu Arg Phe Leu Leu Ser Asn Ala Arg Tyr Trp Leu Asp
385 390 395 400

Glu Phe Lys Phe Asp Gly Phe Arg Phe Asp Gly Val Thr Ser Met Leu
405 410 415

Tyr Thr His His Gly Leu Asn Met Ser Phe Thr Gly Ser Tyr Lys Glu
420 425 430

Tyr Phe Gly Leu Ala Thr Asp Val Asp Ala Val Val Tyr Leu Met Leu
435 440 445

Ala Asn Asp Leu Ile His Gly Leu Xaa Pro Glu Ala Val Val Val Gly
450 455 460

Glu Asp Val Ser Gly Met Pro Val Leu Cys Xaa Pro Val Asp Glu Gly
465 470 475 480

Gly Val Gly Phe Asp Tyr Arg Leu Ala Met Ala Val Ala Asp Lys Trp
485 490 495

Ile Asp Leu Leu Lys Asn Lys Asp Asp Xaa Trp Ser Met Gly Xaa Ile

```
                500                     505                     510

Val His Thr Leu Thr Asn Arg Arg Tyr Pro Glu Lys Cys Val Ala Tyr
        515                     520                     525

Ala Glu Ser His Asp Gln Ala Leu Val Gly Asp Lys Thr Ile Ala Phe
    530                     535                     540

Leu Leu Met Asp Lys Asp Met Tyr Asp Gly Met Ala Leu Xaa Xaa Pro
545                     550                     555                     560

Ser Ser Pro Thr Ile Asp Arg Gly Ile Ala Leu Gln Lys Met Ile His
                565                     570                     575

Leu Ile Thr Met Gly Leu Gly Gly Asp Gly Tyr Leu Asn Phe Met Gly
            580                     585                     590

Asn Glu Phe Gly His Pro Glu Trp Ile Asp Phe Pro Arg Gly Pro Gln
        595                     600                     605

Leu Pro Thr Gly Lys Pro Gly Asn Asn Asn Ser Tyr Asp Lys Cys Arg
    610                     615                     620

Arg Arg Phe Asp Leu Gly Asp Ala Asp Phe Leu Arg Tyr His Gly Met
625                     630                     635                     640

Asn Ala Phe Asp Gln Ala Met Gln His Leu Glu Asp Lys Tyr Gly Phe
                645                     650                     655

Leu Ser Ser Ser His Gln Tyr Val Ser Arg Lys Asn Glu Glu Asp Lys
            660                     665                     670

Val Ile Val Phe Glu Lys Gly Asp Leu Val Phe Val Phe Asn Phe His
        675                     680                     685

Trp Ser Asn Ser Tyr Phe Asp Tyr Arg Val Gly Cys Xaa Xaa Pro Gly
    690                     695                     700

Lys Tyr Lys Val Ala Leu Asp Ser Asp Ala Xaa Leu Phe Gly Gly Phe
705                     710                     715                     720

Gly Arg Xaa Xaa His Asp Xaa Asp His Phe Thr Ser Glu Xaa Xaa His
                725                     730                     735

Asp Asn Arg Pro Xaa Ser Phe Ser Val Leu Thr Pro Ser Arg Thr Cys
            740                     745                     750

Val Val Tyr Ala Pro Xaa Glu Xaa Ala Ala Xaa Val Thr Lys Xaa Tyr
```

755 760 765

Xaa Xaa Xaa Xaa Xaa Leu Xaa Arg Xaa Xaa Gly Xaa Xaa Xaa Xaa Xaa
770 775 780

Xaa Phe Leu Xaa Pro Xaa Lys Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Leu
785 790 795 800

Xaa Xaa Xaa Xaa Xaa Xaa Pro Xaa Xaa Xaa Pro Xaa Ile Xaa Phe Xaa
805 810 815

Xaa Xaa Gly Gly Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Lys Xaa
820 825 830

Xaa Xaa Xaa Ala Val Xaa Xaa Xaa Xaa Xaa Ser Xaa Xaa Xaa Xaa Xaa
835 840 845

Xaa Ile Leu Xaa Leu Xaa Xaa Xaa Xaa Ile Ile Xaa Xaa Xaa Xaa Xaa
850 855 860

Xaa Xaa Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa
865 870 875 880

Lys Lys Lys Lys Lys Lys Lys Lys
885

<210> 55
<211> 1488
<212> DNA
<213> Zea mays

<400> 55
aagcttgcat gcctgcaggt cgactctaga ccaaatttca tggtagttgg gagcctaccc 60
agatttcatg attaactgtg ctattgaatt gttgaaaatg gttgtgtctg tcgtatccga 120
cggataacgg aaacccgtcc gaaattcaat gggcatgggc atagatatag atttgtaccc 180
actactagta tggtcgcagg cggatattgg ttgcaaccgc agatatagtt tcggggaaaa 240
ggattaggct cagctccatc cctagacccc acttgtgtgt gtggggggggt ctacccttca 300
aaaggaaaaa aaactacaca cagtgcatat aagaagatga atattccaaa attcagcagt 360
caagaagccc tgataaactg tctggcatag ctagtacttt atacacttca agaccaaaag 420
aaatcactaa gtacagattt tagtgactcg taagtacaga tatcatctta caaggcccag 480
cccagcgacc tattacacag cccgctcggg cccgcgacgt cgggacacat cttcttcccc 540
cttttggtga agctctgctc gcagctgtcc ggctgcttgg acgttcgtgt ggcagattca 600
tctgtcgtct cgtctcctgt gcttcctggg tagcttgtgc agtggagctg acatggtctg 660
agcaggctta aaatttgctc gtagacgagg agtaccagca cagcacgttg cggatttctc 720
tgcctgtgaa gtgcaacgtc taggattgtc acacgccttg gtcgcgtcga tgcggtggtg 780
agcagagcag caacagctgg gcggcccaaa gttggcttcc gtgtcttcgt cgtacgtacg 840
cgcgcgccgg ggacacgcag agagcggaga gcgagccgtg cacgggggag gtggtgtgca 900

```
agtgcagccg cgcgcccgcg cccgcgcccg gtgggcaacc caaaaagtac ccacgacaag 960
cgaaggcgcc aaagcgatcc aagctccgga acgcatcagc cacaagcagc cgagaaccga 1020
accggtgggc gacgcgtcgt gggacggacg cgggcgacgc ttccaaacgg ggccacgtac 1080
gccggcgtgt gcgtgcgtgc gtgcagacga caagccaagg cgaggcagcc cccgatcggg 1140
aaagcgtttt gggcgcgagc gctggcgtgc gggtcagtcg ctggtgcgca gtgccggggg 1200
gaacgggtat cgtgggggc gcggcggaga agagcgtggc gaggccgaga gcagcgcgcg 1260
gccgggtcac gcaacgcgcc ccacgtactg ccctccccct ccgcgcgcgc tagaaatacc 1320
gaggcctgga ccgggggccc ccccgtcaca tccatccatc gaccgatcga tcgccacagc 1380
caacaccacc cgccgaggcg acgcgacagc cgccaggagg aaggaataaa ctcactgcca 1440
gccagtgaag ggggagaagt gtactgctcc gtcgactcta gaggatcc 1488
```

INTERNATIONAL SEARCH REPORT

onal Application No
PCT/GB 99/03011

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C12N15/82 C12N9/10 A23L1/0522 A01H5/00

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C12N A23L A01H

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 97 22703 A (DU PONT ;HUBBARD NATALIE LOUISE (US); KLEIN THEODORE MITCHELL (US)) 26 June 1997 (1997-06-26) cited in the application	1-4, 6-15, 17-25
Y	see the claims see SEQ ID NO: 1 (page 50-53) abstract; figures 1,2,6-15; examples 1-3,7 page 1 -page 7 page 14, line 29 -page 21, line 35	14-25

-/--

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
20 December 1999	11/01/2000

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Oderwald, H

2

nal Application No
PCT/GB 99/03011

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
X	NAIR R B ET AL: "Isolation, characterization and expression analysis of a starch branching enzyme II cDNA from wheat" PLANT SCIENCE, vol. 122, 1997, pages 153-163, XP002095263 cited in the application	5,7, 9-14, 16-25
Y	abstract; figures 2-5 page 154 -page 156 page 159 page 162	15
X	SUN C. ET AL.: "The two genes encoding starch-branching enzymes IIa and IIb are differentially expressed in barley" PLANT PHYSIOLOGY, vol. 118, 1 September 1998 (1998-09-01), pages 37-49, XP002095264	1-13
Y	abstract; figures 1-3 page 45, paragraph 7 -page 47, paragraph 2	14-25
P,X	WO 99 14314 A (GOODMAN FIELDER LTD ;LI ZHONGYI (AU); MORELL MATTHEW (AU); RAHMAN) 25 March 1999 (1999-03-25) abstract; claims 1-52 see SEQ ID NO: 10 and 12 (pp.75-81 and 83-85) page 6 -page 10	5-7,9-25

2

INTERNATIONAL SEARCH REPORT

International application No.
PCT/GB 99/03011

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

See additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. ☒ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International Application No. PCT/ GB 99 /03011

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: 1-4,6 8, all complete; 7, 9-25 all partially

   Nucleotide sequence encoding wheat SBEII-1 members 5A1, B2, B4, B10 or B6 3' UTR sequences thereof, vectors, host cells, amino acid sequences encoded by said nucleotide plant; plants and parts thereof, starch, a method for making altered starch, use of that starch, foodstuff containing said nucelotide sequences.

2. Claims: 5 complete; 7, 9-25 partially

   Nucleotide sequence encoding wheat SBEII-2 member B1. Vectors, host cells, amino acid sequence encoded by said nucleotide sequence. Methods for altering the chracteristics of a plant; plants and parts thereof starch, a method for making altered search, use of that starch, foodstuff containing said starch using said nucelotide sequence.

INTERNATIONAL SEARCH REPORT

tion on patent family members

nal Application No
PCT/GB 99/03011

Patent document cited in search report		Publication date	Patent family member(s)			Publication dat
WO 9722703	A	26-06-1997	AU	1684697	A	14-07-1997
			BR	9612086	A	17-02-1999
			CA	2239979	A	26-06-1997
			CN	1219199	A	09-06-1999
			EP	0868520	A	07-10-1998
			HU	9902112	A	28-10-1999
WO 9914314	A	25-03-1999	AU	8967098	A	05-04-1999